POSTHARVEST PHYSIOLOGY OF POMACEOUS FRUITS

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INTRODUCTION

Pomaceous fruits like other fruits continue to live after they are removed from the trees. Thus they carry on different physiological processes characteristic of living organisms. The physical processes are accompanied by certain chemical changes. Therefore, a knowledge of the physiology and chemical change of these fruits after harvest is useful in understanding what happens to the fruits in storage, in the marketing channels, and in the hands of the consumer.

Many attempts have been made to review the great amount of literature on postharvest physiology that takes place after various kinds of fruits are harvested. Pertinent findings have been published in review by Biale (7). Smock (132) has written on the postharvest physiology of deciduous fruits in storage. Miller (109) has written on the physiology of citrus fruits in storage, and Pentzer and Heinze (119) on the postharvest physiology of fruits and vegetables, with particular emphasis on the role of volatile emanations in certain physiological diseases. Magness et al. (98), Hawkins (53), Brooks (12), and Wright (164) have referred to carbohydrate transformation in fruits and vegetables. Nitsch (114) wrote on fruit growth and problems of metabolism, and Ulrich (152) on the postharvest physiology of fruits, with special reference to the conditions of ripening, the gas exchanges, and the metabolism of postharvest fruits. Much information may also be found relating to the postharvest physiology

of apples, lemons, and cherries in the books of Smock and Neubert (138), Bartholomew and Sinclair (6), and Marshall (99).

The purpose of this report is to review the literature pertaining to postharvest physiology of pomaceous fruits with special reference to apples and pears.

THE CONDITIONS OF RIPENING

Several studies have been published indicating that nutritional and climatic conditions during the life of the plant have important effects on the postharvest behavior of fruits. It has been found that many physical or chemical agents may have significant effects on ripening of the fruit after picking.

1. Temperature:

When fruits are picked at the right time they generally can ripen at any temperature between two critical limits. Leonard et al. (95) reported that Bartlett pears stored at 34°F continued ripening but at a much slower rate than at 70°F. Temperature does not affect the development of color and firmness at the same rate. Certain varieties need cold treatment before ripening at high temperatures. Passe Crassane pears when held at 32°F for 11 to 15 weeks ripen well at 18°C. Ethylene has the same effect as cold pretreatment. According to Childers (20) fall and winter varieties of pear attain the highest quality by storing in rooms at 30 to 31°F and ripening at temperatures between 60° and 70°F. Fisher and Porrit (33) reported that Bartlett pears harvested one and

two weeks beyond the date of commercial harvest ripened satisfactorily with good quality stored at 31° to 32°F for six weeks. A slight change in temperature affected the maturity to a considerable extent.

According to Childers (20) ripening processes of apples proceed slowly when the temperature is maintained at 32°F. The ripening rate of apples is approximately doubled or tripled for every 18°F rise in temperature. Haller and Lutz (46) reported that the rate of softening of apples in storage depends upon the temperature at which they are stored. For a number of varieties the rate of softening at 40°F was found to be slightly more than double that at 32°F, when stored at 50°F was slightly less than double than at 40°F, and that at 60°F was nearly double the rate as 50°F.

They concluded that softening was due to the conversion of an unsoluble specific substance such as protopectin, into a soluble form. They found that the rate of conversion at different temperatures was proportional to the rate of softening.

2. Radiations:

Morgan (111) stated that radiations may act as stimulators or inhibitors of ripening. Loss of texture and some loss of color and flavor during irradiation are likely to be common in the case of many fruits. Complications are often introduced by the persistence, after irradiation, of metabolic activity of the product or activity of individual enzymes. Apples develop a pronounced 'irradiation' flavor with a dose of 2,000,000 rep. (roentgen-equivalent-physical = the energy lost by fast electrons in producing, in air, ions of either sign carrying one electro-static unit of charge). Apples and pears may lose flavor at 2,000,000 and 4,000,000 rep.

with no obvious loss of texture and if allowed to stand overnight may become brown, as though bruised. According to Huber and co-workers (56) oxygen is one of the principle factors responsible for radiation initiated organoleptic changes.

3. Humidity:

Maturity of fruit, especially the evolution of the flavor, is influenced by the relative humidity and the velocity of air in the vicinity of the fruit. The development of a good flavor or odor in pears may be hindered by saturated air; apples may show blackening of the core under similar conditions, Ulrich (151). According to Childers (20) apples should be stored at 85 percent and pears between 80 and 85 percent relative humidity.

4. Ethylene:

Childers (20) stated that ethylene is the most important of the chemicals used to effect ripening. It acts on the ripening of the various fruits, it hastens the ripening of preclimacteric pears, except for the Kieffer variety. Its effects on the ripening of pears are apparent only if the gas is used a short time after harvest. If the pears have been in cold storage for several weeks, they show little or no response to it. Its effects are most pronounced at temperatures of 60°F to 70°F with little or no effect at cold storage temperatures. Ulrich (151) reported that the effectiveness of its treatment on ripening at ordinary temperature decreases when the preliminary cold storage period is increased. Its effects on respiration and ripening are limited at low temperatures.

Biale and co-workers (8) suggested that where ripening of fruit is

uniform without ethylene treatment, use of this gas has no advantages. Native ethylene is a product of the ripening process rather than a casual agent.

Amount of ethylene required to stimulate ripening:

There is a close relationship between ripening and respiration of fruit. In order for ethylene to stimulate respiration and ripening of fruit in storage ethylene must reach a certain minimum concentration or threshold value which must be maintained for a certain period of time. While the presence of ethylene is associated with climacteric rise and onset of ripening, unless other conditions are suitable for ripening, ethylene has little effect. Hansen (47) suggested that the specific reaction in which ethylene is formed must first occur in order for fruit to ripen: Hansen (48) also stated that the small quantity of ethylene arising from immature Bartlett and Anjou pears, about 1 part in 22 million, produced leaf epinasty but had no effect upon respiration of fruit. However, a concentration of 1:1000 caused immediate respiratory stimulation.

5. Volatiles:

According to Smock (132) no aspect of postharvest physiology of fruit is as controversial as that of the role of fruit volatiles. Different results have been found by different investigators who worked on this problem. However, Smock found that non-ethylenic volatiles may be influential in ripening apples at storage temperatures. The vapors of ripe apples aspirated over preclimacteric apples made them respire and soften faster. Kidd and West (84) showed that volatiles produced by one lot of apples could increase the ripening rate of another lot. Maxie and Baker (104) reported

that inclusion of post-climacteric fruit as fillers stimulates the ripening rate of pre-climacteric apples in the same room. Smock and Gross (136) reported that ripe apricot vapors have effects on the ripening of immature apricots, pears and apples. However, it was found that the stimulation in ripening was not proportional to the amount of apricots used. Volatiles from large quantity of ripe apricots had a less stimulatory effect on the ripening of these fruits than from smaller quantities at high temperatures. When activated carbon was used in the recirculation system, it tended to remove the depressant effect of a large quantity of apricots. This indicated that a depressing volatile was involved. Smock (133) in a separate study found that relatively few ripe apples had more stimulatory effect than many at high temperatures.

Ulrich (151) reported that the presence of ripe fruits under certain conditions has been found stimulating to unripe fruits. However, this has not been confirmed with apples and pears stored at -1°C. Air purification had no influence on apples at -0.5°C.

The ripening of preclimacteric fruits in cold storage which can be stimulated by the volatiles of ripe fruits is a problem which still has to be solved. Different workers using different conditions of temperature, and different varieties and species in their experiments have found conflicting results. According to Grierson-Jackson (38), non-ethylenic volatiles, in a cold room, may condense on the evaporator coils in considerable quantities.

6. Growth substances:

Growth substances are used to stimulate the ripening of harvested

fruits. Their treatment is only effect under certain conditions, particularly when they are used very early after picking, as indicated by Ulrich (152). Mitchell and Marth (110) used 2,4-dichlorophenoxyacetic acid to test its effect on the ripening of different varieties of apples after picking. They reported that yellow Newton fruit ripened during the two weeks period immediately following treatment where untreated apples failed to ripen in this length of time. Grimes Golden apples ripened within a period of 6 days, where untreated apples took two weeks at room temperature to reach the same stage of ripeness. Rome Beauty apples ripened two to six days earlier than the untreated ones. Kieffer pears took two to four days less than required for comparable untreated fruits. Winter Bartlett pears ripened within a period of eight to ten days following treatment, while untreated fruits failed to ripen during this period. Treated fruits of both varieties of pears ripened more uniformly than the control.

In studying the influence of α-naphthaleneacetic acid spray on the maturity and storage physiology of apples and pears, Gerhard and Allmendinger (36) found that this mormone had no influence on the quantity and degree of ripeness when those fruits were harvested within their normal range of acceptable picking maturity from one to two weeks after application of the spray. It was further found that when these conditions were ignored, the rate of ripening was increased to the extent that losses from breakdown became serious.

Smock and Gross (136) reported the effect of various hormone materials on apples when application was made prior to harvest. Southwick (141)

showed that 2,4-D and methyl α-naphthalene acetate were effective in stimulating the ripening of peaches and apples. Hansen (50) reported effects of 2,4D and accumulated volatiles on ripening of immature and mature Bartlett pears.

GAS EXCHANGE

Pathway of gas exchange:

Ulrich (152) reported that after the fruits have been harvested they take in oxygen, water vapor or carbon dioxide and give out carbon dioxide, ethylene, volatiles and water vapors. The internal atmosphere surrounding the living cells of the fruits does not have the same composition as air. Therefore, the gas exchanges between the living cells of the fruits and the air are, for the main part, indirect. The circulation may take place through intecellular spaces, the lenticels, the skin, calyx, and superficial wounds.

Pathways of circulations are not the same for different varieties and sometimes in fruits of the same variety. There may be differences from one fruit to another. The calyx, sometimes, becomes an important pathway for the circulation of gases. With the presence of a number of functional lenticels, or when large wounds are present, the permeability of apples increases with the pressure of the air, and decreases during ripening and senescence. Only a little part of the water vapor and carbon dioxide evolves through the lenticels; most diffuses through the cuticle. Oxygen, which is less soluble in water and in lipids than carbon dioxide, enters

through lenticels.

Reeve (124) reported that changes in gas composition accompany other changes related to structure and texture in stored apples. The gas content increases while specific gravity decreases with mealiness and maturity in apples. Trout, Hall and Sykes (149) reported that the keeping quality of apples and of certain fruits may be improved considerably by a suitable skin coating. The effects of the coating are found to depend greatly on temperature, thickness and type of coating, and variety and condition of fruits. Coating increased the resistance of the skin to the gaseous diffusion and thus greatly reduced the internal oxygen concentration, increased the internal carbon dioxide concentration, reduced the respiration rate and retarded ripening changes by varying degrees, and caused a marked retardation of normal yellowing of the skin which is mainly controlled by internal oxygen supply. A marked change in the composition of gas in the fruit tissues under varying conditions of temperature was reported by Magness (97).

Respiratory gas exchanges:

Claypool, Maxie and Esau (22) found that the rate of air flow over respiring fruits may be a critical factor in carbon dioxide production. Two phenomen may be involved - the retarding effect of carbon dioxide accumulation or oxygen depletion, and the stimulatory action of volatiles. Ulrich (152) reported that carbon dioxide output is not always a good test of fruit ripening. The rate of ripening of plums is not increased by high temperatures in the same proportion as carbon dioxide production, and a peak of carbon dioxide may be observed prior to ripening in tomatoes.

The Climacteric:

Biale (7) reported that the respiration of fruit, measured by the cells' uptake of oxygen, is high during the stage of cell division and gradually decreases during enlargement and maturation of the cells. After the cells mature, there comes a sharp rise in respiration rate followed by a decline. This phase of respiration has been named the "climacteric." Temperature has a marked effect on the course of respiration during the climacteric stage. In certain fruits, the higher the temperature, the sharper the rise and higher the peak. Low temperatures tend to suppress or completely obliterate the climacteric.

Biale and his co-workers (8) found the occurrence of the climacteric rise in carbon dioxide production in several species of fruits. The fruits with a marked climacteric showed high rates of ethylene production. Mango is an exception. The ratio of ethylene evolution to carbon dioxide output was highest for the apple, followed by the sapote and pear. In practically all cases ethylene was found to accelerate the onset of the climacteric if applied before the rise.

The information available on the relationship between ethylene production and respiration is limited and conflicting. Nelson (113) found that the sharp rise in ethylene production of apples followed the rise in carbon dioxide evolution. In the case of bananas an inverse relationship between ethylene and carbon dioxide evolution has been observed. Hansen (47) working with pears observed that the maxima in both processes occurred at the same time.

Hulme (64) followed over a number of seasons the respiration rate of

apples kept at 12°C and 15°C. He noted that there is a regular biennial variation in the level of respiration, and the difference in the time between the onset of the climacteric at 12°C and 15°C varies with the date of picking. He further found that the ratio of rate of respiration to protein content at the climacteric peak is constant within one variety.

Effects of External Conditions and of Chemicals on Respiratory Activity:

Ulrich (52) reported that the effect of temperature on respiration in apples is not the same for the skin and for the pulp, and that it differs with the seasons. At low temperatures the CO_2/O_2 ratio of oranges increases; thus respiration becomes more dependent on utilization of organic acids. It may explain the cause of the poorer taste of fruits stored at low temperatures. Carbon dioxide production by apples and pears placed in wet air on in air circulating at a high speed may be higher than that of fruits placed in opposite conditions.

Claypool et al. (21) found that plum fruits held in oxygen levels above that in air are accelerated both in ripening and respiration rates. The rate of acceleration was proportional to the oxygen tension. Caldwell (17) showed that partial pressures of the order of 500 percent oxygen are toxic to the tissues of apples and that there is no evidence of an initial increase in carbon dioxide output over that in one atmosphere 100 percent oxygen. It was further found that the actual pressure (up to 20 atm) is not responsible for the disorganization of the cells but that there is specific oxygen toxicity. Ulrich (152) reported that the optimum in respiratory activity obtained for a certain concentration of oxygen is not the same for young and old apples or for apples stored at 10°C and 30°C.

Fidler (30) found that the respiration of apples is depressed in gas storage at 4°C. A respiration peak was found which is similar to the peak of the climacteric rise in air, but he believed that it was not the climacteric. The immediate effect of high concentrations of carbon dioxide in the gas-storage of apples is to induce a transitory rise in the rate of respiration. Following this, it depresses respiratory activity below the level of normal respiration in air.

Ulrich (152) reported that ripening and the climacteric rise can be prematurely induced at 20°C by giving the fruit ethylene at low concentration. But it seems that at 3°C the respiration is not sensitive to ethylene. At 7°C the effect is slight but it becomes marked at 12°C. Griffiths and Potter (41) found that the continuous addition of ethylene, between 20 and 500 part per million, had little effect upon the respiratory activity of apples stored at 5°C in a continually renewed gas mixture. Fidler (32) expressed an opinion that the effect of ethylene on respiration is due to an uncoupling of phosphorylation, the reason for this uncoupling is unknown.

Uota and Dewey (154) found that postharvest applications of 2,4,5-T (200 ppm) and ethylene (20-30 ppm) at 70°F are effective in accelerating the rate of respiration of pre-climacteric Bartlett pears. The effects of both compounds are greatly reduced when applied to the fruit at 70°F following a 15 day period of cold storage at 35°F. Many authors have found that the respiration of matured picked lemons can be stimulated by a mixture of ozone and 1-hexene. In the case of apples the peel is the region showing the highest respiratory activity, this observation is perhaps correlated with the fact that the peel is the tissue where cell divisions

continue longest.

Volatile Organic Products of Metabolism of Fruits:

Non-ethylenic Volatiles:

The non-ethylenic fraction (the odorous volatile substances) is believed by many to contain substances which have physiological effects and can be troublesome in storage chambers. The following methods are used for the collection of volatiles:

1. Distillation from fruit tissue or juice (158).
2. Absorption on activated carbon from the air of a fruit store (93).
3. Collection of the products in reagent solutions or cold traps from air sent over fruits (146).
4. Steam distillation at atmospheric pressure.

For identification of volatiles new methods have been investigated including use of ultraviolet and infrared spectra, and chromatography after preparing suitable derivatives.

Composition of the Volatile Mixture:

Results were obtained from the work done on apples. The following components have been reported:

1 (a). The chemical composition of the aroma of apples was investigated by Jonathan (76). He found that the principal components, together with the relative amounts of each class, are alcohols (92 percent): methanol, ethyl alcohol, propyl alcohol, 2-propanol, butyl alcohol, isobutyl alcohol, d-2-methyl-1-butanol, and lunyl alcohol; carbonyl compounds (6 percent): acetaldehyde, acetone, caproaldehyde, and 2-hexanol esters (2 percent): ethyl

butyrate and ethyl caproate. Menthanol, ethyl alcohol, 2-propanol, butyl alcohol, and formic, acetic, propionic, butyric and caproic acids were identified as components of other esters. These components are present in the original apple juice at a total concentration of approximately 50 ppm.

1 (b). White (158), Thompson (146), Heinze et al. (55), and Meigh (109) reported the following components: Alcohols: menthanol, ethanol, N-and isopropanol, N-and isobutanol, d-2-methyl-1-butanol, and hexanol; Esters: esters of methanol, ethanol, 2-propanol, butanol, hexenol, and formic, acetic, propionic, butyric, caproic, and valeric acids; Aldehydes and Ketones: aldehyde, 2-hexanol, caproaldehyde acetone, n-butanal, propanal, ethylmethyl-ketone, isobutanal, with probably isovaleraldehyde and methylpropyl ketone in traces and other carbonyl compounds. Some of these substances are responsible for the odor and flavor of the ripe fruits.

Thompson and Huelin (147) found that, in every removal of apples at intervals from 0°C to 20°C, ester production at 20°C increased to a maximum and then decreased; in later removals the increase was much less and finally negligible. At 0°C ester production increased steadily. A higher rate of air flow increased it and reducing the oxygen concentration to 6 percent first increased then decreased in comparison with air.

There has been a general agreement that acetaldehyde is the most abundantly produced carbonyl compound at ordinary temperature. At cold storage temperature, however, Heinze et al. (55) found mostly acetone, and in Meigh's (109) work the main contributor of the volatile matter with apples was acetone, with smaller amounts of acetaldehyde, n-butanal, propanal, ethyl methyl ketone and isobutanal, with isovaleraldehyde and

methypropyl ketone in traces.

Heinze et al. (55) reported that the influence of ripeness of fresh Bartlett pears on flavor and on the quantity of various volatile reducing substances in the canned product has been investigated. It has been found that as pears ripen at 68°F (20°C), methyl alcohol, total carbonyl compounds, acetyl methyl carbinol, diacetyl and ester content gradually increases. The rate of production of such compounds is especially rapid when the pressure test of the fresh pears drops below 2 pounds. The presence of excess methyl alcohol in overripe Bartlett pears is thought to be caused by de-esterification of the methyl ester group of the pectin molecule by the enzyme pectin esterase.

Ethylene:

It has been found that the complex metabolic processes of fruit during growth and after abscission produce many compounds, mostly organic both solid and gaseous. Of these, ethylene has the most practical significance and has been the subject of much research. Its chemical identification among the volatile products of ripening fruits has been accomplished in apple, banana, pear, and avocado.

It is an unsaturated hydrocarbon gas, non-poisnous, with a faint sweetish odor. Its boiling point is -103.9°C and specific gravity 0.97g compared to air. It is soluble in water to the extent of 25.6cc per 100 gm of water at 0°C.

There is evidence that presence of this gas in storage atmosphere tends to accelerate the ripening process in fruit. It is also known that fruits themselves generate ethylene. Certain metabolic processes may be

accelerated by this gas. Thus, normal reactions such as destruction of chlorophyll, conversion of starch to sugar, hydrolysis of sucrose to reducing sugars and hydrotysis of protopectin to soluble pectins may be stimulated by ethane. No changes, however, are produced by ethylene treatment that would not normally occur during the regular course of ripening. It has also been found that its treatments do not effect a change in edible portion of fruits and vegetables.

Methods of Identification:

Different methods and processes have been developed for identification and determination of this gas. Among them are:

1. Estimation of ethylene by paper sensitizer by red selenium to mercury vapor (144).
2. Manometric method for determination of low concentration of ethylene, involving the production of a complex with mercuric perchlorate (162).
3. Perchlorate fixation method plus oxidation with ceric salt (153).
4. An absorption technique using mercuric perchlorate combined with the use of a Katharometer.
5. A quantitative microbromination method (23).

Ethylene Production:

The production of ethylene varies between different species and between the varieties of the same species. Variations of its production during the ripening period are particularly pronounced and bear relation to respiration and to storage life. Biale et al. (8) concluded that ethylene is a product of the ripening process rather than a causual agent.

Uta (153) showed that ethylene is produced by plums under conditions favorable to ripening. Gerhardt (35) working on pear and apples reported that Bartlett pears produced a maximum of 0.87 mg of ethylene per Kg per day after 84 days at -1°C; this was more than 14 times the rate of Anjou variety. The maximum rate from apples during cold storage usually varied from 0.3 to 0.54 mg. Emanation was highest as 18°C.

Many fruits from tropical and temperate climates have been investigated by Biale et al. (8). Species showing marked climacteric give off ethylene, with the exception of the mango. The ratio of ethylene evolution to carbon dioxide output is highest for the apples, followed by the pear and the peach. Oranges and lemon do not exhibit any climacteric nor do they produce ethylene.

The very young fruits (pears, cherries) do not seem to produce ethylene. At the end of their growth, the quantity evolved is zero for cherries and 0.7 and 1.8 cc/kg/day for Williams pears, 2 cc/kg/day for Canada apples and 11 cc/kg/day for Reine apples (152).

Hansen (47) reported that each pear variety has a characteristic maximum rate of ethylene production which varies but little in different lots. Fidler (30) stated that apples of the same variety, but grown in the different districts may produce ethylene at different rates. In general, Fidler listed dessert apples such as Cox orange as producing more volatiles than culinary varieties of the Bramley seedling clan, which incidentally have longer storage life. Nelson (112) and Hansen et al. (51) found that varieties of apples with long storage life have less ability to produce ethylene than short life varieties.

Hansen (49) also found that an inverse correlation exists between length of maturity period of apples and the amount of ethylene produced during ripening. Thus Astrachan and Red June mature early and have a maximum ethylene output of 11 and 9 ml/kgm/24 hrs., respectively, while Delicious and Newton evolve about 1.7 ml of ethylene for the same period.

Gane (34) found the total production of ethylene during postharvest life of an apple to be approximately 1 ml at 20°C. Hansen (47) found a varietal difference of 1 to 4 ml of ethylene per bu/day, depending on variety and the length of time in storage.

Relation of Ethylene Production to Respiration:

Biale (7) reported that from the standpoint of storage life of fruit the dominant metabolic activity is respiration. The rate of respiratory activity, as measured by oxygen - carbon dioxide exchange, is an index to the rate of metabolism and hence the length of life of fruit. Factors which are associated with the rise in the respiratory activity appear to be similar to those associated with volatile production. These two functions follow the same general trends throughout the storage life of the fruit; however, the peaks do not always correspond. As fruit matures and ripens, ethylene production increases to a peak, concomitant with the respiratory climacteric and then declines as does the respiratory activity.

Nelson (113) found that the production trends of ethylene and carbon dioxide in apples and bananas are very similar. The ethylene output increases during the respiratory climacteric, declines rapidly, rises again briefly, then subsides slowly during senescence. He also stated

that the peak of ethylene production in McIntosh and several other varieties of apples, lags behind the respiratory peak by about five days at 20°C. Hansen (47) reported that the ethylene production in pears follows a somewhat different trend than apples and bananas. The peak in ethylene output occurs about the same time as the respiratory peak, and during the senescent decline no further rise occurs to correspond with the brief respiratory rise.

Hansen (48) also made a detailed quantitative study of ethylene production in relation to respiration of pears at different temperatures and under different conditions of oxygen tension. Bartlett pears which ripened immediately after picking showed an increase in respiration and ethylene production about the same time and the maxima in both processes occurred at the same time.

Although ripening and the occurrence of a respiratory climacteric appear to depend upon a supply of ethylene which also increases during this period, the two processes may not be directly correlated. Hansen (47) has given evidence for this in the comparison of carbon dioxide and ethylene production. During the climeteric rise there is a much greater increase in ethylene than in carbon dioxide production.

Hansen (47) stated that the comparison of the emission curves of carbon dioxide and ethylene during ripening together with the fact that ethylene not evolved under anaerobic conditions lead to the idea of a link between respiration and ethylene formation. However, respiratory activity and ethylene production do not follow the same trend in a range of temperature from 0°C to 40°C. At 40°C the carbon dioxide of respiration is at a

maximum while the ethylene is at minimum. On the other hand, Ulrich (152) reported that a concentration of DNP insufficient to cause a strong inhibition of respiration can stop ethylene production completely. According to Hall (44), ethylene may act as an autocatalyst, accelerating its own production, in respiring fruits.

Total Production of Volatiles:

Fidler (32) reported that the loss of carbon in the form of volatile organic substances probably never exceeds 1 percent of that lost as carbon dioxide. For apples, which are the fruits to be considered in most detail, the figure is about 0.1 - 0.3 percent. Cooking apples produce about 0.2g of organic volatile substances/ton/day at 3°C. Dessert apples produce about 3.5g/ton/day at 0°C. In terms of carbon, ethylene forms 70-80 percent of the total. The remainder is usually referred to as the "odorous fraction."

The Effect of Temperature on Respiration and Volatile Production:

The effects of temperature changes on fruit metabolism, although more complex, are similar to those on other chemical processes. Raising the temperature of fruit increases the respiratory activity, causing a corresponding increase in O_2-CO_2 exchange and decrease in respirable substrate. Raising the temperature also induces a corresponding increase in production of volatile materials. According to Southwick (140) McIntosh apples produce twelve times more volatile materials at 40°F than at 32°F; while at 74°F apples emitted as much organic vapors in one day as in five months at 32°F.

Variations in temperature have different effects on respiration and ethylene production. Hansen (47) investigated the effect of temperature on

respiration and ethylene production. Between 0°C and 20°C both processes increased; from 20°C to 40°C carbon dioxide evolution continued to increase steadily but the rate of ethylene production declined sharply, reaching a zero value at 40°C. The rate of soluble pectin formation paralled the ethylene picture. The suggestion was made that at higher temperatures oxygen concentration might be limiting ethylene production, since it was found that the oxygen content of the tissue decreased sharply with increasing temperature.

According to Uota and Dewey (154) the production of volatiles by pears is affected by ethylene and 2,4,5-T in about the same way as carbon dioxide production, but ethylene does not affect the volatile production when applied at 2°C.

The Effect of the Oxygen Content of Storage Atmosphere:

With low oxygen supply, ethylene production is greatly repressed or entirely inhibited, but carbon dioxide production may continue in either aerobic or anaerobic condition, at least to some extent. Not only does low oxygen tend to inhibit the production of ethylene, but, according to Kidd and West (88), the ripening effect of ethylene diminishes with reduction of oxygen, unit at 0.3 percent or less the ethylene has no effect on respiration. Thus the principle of "controlled atmosphere storage" or "gas storage", involving low oxygen and relatively high carbon dioxide atmospheres, functions to depress ethylene production and delay the climacteric rise, thus reducing the ripening rate. Storage life of individuals in any one variety under similar storage conditions may vary, being influenced by maturity, size and growing conditions. Coincident

with long life is low respiratory activity, small size, prime maturity, and a small and an extended climacteric.

Mattus (103) reported that after cold storage, pears taken from a controlled atmosphere have lower rates of production of carbon dioxide, ethylene and other volatiles than fruits taken from air storage. In the same way Fidler (31) found that the rate of production of ethylene and non-ethylenic volatiles from apples at 4°C in air to be higher than in a gas storage. However, according to Hansen (47), increased oxygen in the atmosphere surrounding the fruit does not result in increased ethylene production at high temperatures.

Functional Diseases and Volatile Emanations:

Postharvest functional diseases of fruits are manifestation of different causes:

1. Chilling injury - Soft scald, internal browning, or brown core of apples, breakdown or browning of the flesh of peaches, plums, avocados, pineapples, skin pitting of citrus fruits; and certain types of internal breakdown of apples and pears that appear in overmature fruit or fruit held too long in storage fall into this group.
2. Lack of oxygen - Brown heart of apple is a well-known disorder of this type.
3. Volatile emanation - Apple scald is a disorder of apples caused by the accumulation of apple volatiles in the apple skin when apples are stored at low temperatures. The problem of the production of volatiles is very important from a practical point of

view, as it concerns the origin of scald and the effectiveness of air purification by activated carbon or by alkaline permanganate in cold storage.

APPLE SCALD

This problem has caused heavy losses in stored apples and still does in years when apples are very scald susceptible. Numerous experiments have been done to try to understand the origin of apple scald and the means to be used to avoid it.

Studies developed on cause of apple scald:

As early as 1903 Powell and Fulton (123) stated that the disorder is not physiological in origin and is not caused by bacteria or fungi. The brown discoloration of the skin of the apple characteristic of the disease gave it the name "scald" for the skin has a brown, cooked appearance. It was found that fruit picked too early is particularly susceptible to scald, that more scald occurs at 36°F than at 32°F and that the temperature at which the fruit is held when removed from storage has a marked effect on scald development. Varieties of apples are found to differ in susceptibility.

Pentzer and Heinze (119) reported that high humidities favor scald, because it develops in apples held in saturated atmospheres in closed but not sealed containers. They also stated that apples in gas storage at 38°F develop less scald at 80 percent humidity than at 96 to 98 percent. They concluded that the rate of water loss is one of the factors concerned in scald and that conditions favoring water loss would aid in removal of

the volatiles responsible for scald. Brooks and Cooley (13) found that humidity is not important and showed that lack of aeration is the essential factor in scald production. As storage temperature is raised from 32°F scald develops sooner, about a month earlier for each 9°F rise up to 59°F or 68°F. At 86°F no scald develops but breakdown of the flesh occurs.

Further work by Brooks, Cooley and Fisher (14) led them to conclude that they must be dealing with a volatile or gaseous substance other than CO produced by the metabolism of the apple as the causual factor for scald. Since then numerous studies have been made to understand the nature of these volatiles and the way they produce injury to the apple. Gerhardt (35) and Fidler (30) reported that climatic conditions probably also play a role in the development of scald.

Pentzer and Heinze (119) reported that in cold storage some varieties of apples produce a substance which is toxic to the surface cells and ultimately causes death and oxidative browning of the tissue. This disorder is called apple scald. They further stated that total volatile output of apples is not a measure of scald susceptibility. Nonsusceptible varieties may give off more volatiles than the susceptible ones; and early picked fruits, most susceptible to scald, have a lower output of volatile esters than the late picked fruits. Whatever the toxic substances are, they result in death of cells, lowered respiration of affected skin, and enzymatic browning accompanied by decrease in phenolic content.

Fidler (30) developed a new theory about the cause of scald. He stated that scald is caused by two factors, Y, which is volatile and produced later in the season; and X, which is not volatile and produced early in the season.

Factor Y, which is fairly volatile is capable of producing scald only in combination with factor X.

Apple Volatiles Related to Scald:

The question of the cause of scald still remains unsolved. The works reported on volatiles provide evidences of the complexity of the problem of identifying the causual agent or agents of scald among the esters, acids, alcohols, aldehydes, and ketones present in apple volatiles. A considerable amount of work has been done on the chemical nature of apple volatiles (119).

In a study of volatile products of apples in relation to scald, it has been found that Granny Smith apples held at 86°F gave off volatile aldehydes and ketones as well as alcohols and traces of esters. Thompson and Huelin (147) found that early picked fruits gave off a smaller amount of volatile esters than fruits picked later though early picked fruits are more scald susceptible than late picked fruits. They further reported that experiments with synthetic esters failed to provide evidence to support a direct relation between volatile esters and scald. Fidler (30) reported that scald is not directly related to the amount of volatiles the fruits give off, but it does not follow that volatiles or precursors of volatiles are not in some way involved in the disorder.

Griffiths and Potter (40) suggested that the causative conditions for scald may be the accumulation of precursors of the odorous volatiles rather than of the volatiles themselves. Kuc et al. (94) undertook interesting investigations to determine which volatiles might be responsible for apple scald. Crude ester extracts from activated carbon used in commercial

storage are shown to be very effective in providing scaldlike injury. Ethers comprise the most active fraction of the ether extract. Meigh (107) reported that volatiles play little or no role in the development of scald.

Skin Composition and Scald:

The natural coating of the apple skin has attracted attention because it is important in the physiological behavior of the fruit - forming a barrier to the diffusion of volatiles, water vapor, CO_2, oxygen, nitrogen, and other gases. Several studies have been reported by Bentzer and Heinze (119) on the composition of the skin of apple fruit.

Other work on the skin of apples in relation to scald has been done in the U. S. Department of Agriculture laboratories at Wenatchee, Washington. It has been found that the phenolic content of the skin of apples decreases with scald development. Thus, it indicates that the brown color is formed by the action of enzymes on phenolic compounds and in this respect resembles browning of peaches and other fruits. Respiration determinations on incipient - scalded skin and normal skin of apples have been made as parts of these studies. Volatiles from apple storage rooms do not increase the respiration of apple skin. Scalded skin has a lower respiration rate than normal skin. The reduction in respiration is quantitative, indicating that a portion of the cells of the skin are no longer functioning and presumably dead.

Critical period for scald developments:

Brooks, Cooley, and Fisher (14) suggested that there are four stages in the development of apple scald.

1. The first period:

It starts at picking dates and extends for 6 to 8 weeks in storage. During this period, scald-producing agents are most active and scald could be prevented by aeration or use of oiled wraps.

2. The second period:

The next 5 to 8 weeks comprise the second period. Preventive measures are of little avail and the fruit is doomed to scald if left in storage long enough. On the other hand, if the fruit is removed from storage before the end of this period the fruit might not show scald even upon warming.

3. The third period:

The third period is the rest of the cold storage life. Then the fruit is potentially scalded, certain cells get practically dead, but the fruit remains green and appears almost normal if not exposed to warm air.

4. The fourth period:

The fourth period is the life of apples after removal from storage, when the skin turs brown and completion of the death processes take place.

Pentzer and Heinze (119) reported a work carried on with English varieties of apples to determine the critical period of scald indicating that the second 3-week period of storage is most critical for normally harvested fruit and the third and fourth periods of 3 weeks are also critical for immature fruit.

Treatments for Control of the Apple Scald Disease:

The apple scald disease is so serious that it is the factor limiting the storage life of a number of varieties of apples. Several studies have been done on the control of this disease. Since the classic work of Brooks,

Cooley, and Fisher (14) paper impregnated with mineral oil has been the standard method of control. Objections to this method have led to a number of other approaches.

Air purification with activated coconut shell carbon (137) or alkaline permangnate (94) has not given consistently good control of scald. Prestorage treatment of apples with high concentrations of carbon dioxide (142) has often given good scald control, but has sometimes produced secondary undesirable effects. Oil coatings have been tried by several investigators with variable results (15, 135, 140).

Kidd and West (84) reported that intermittent warming of apples every two weeks for 24 hours at 59°F controls scald and warming every 4 weeks gives marked reduction. This gives further support to the belief that accumulation of volatile products at cold storage temperature is the cause of scald. Though it has been presumed that accumulations of volatiles around the apples have been the cause of scald, the exact cause is not known. Fidler (30) has produced evidence to show that volatiles alone were possibly not the cause. Thompson and Huelin (147) and Meigh (108) reported that volatiles play little or no role in development of scald.

Smock (134) concluded that dip treatments of 500-2000 ppm diphenylamine shows promise in controlling apple scald. Wraps impregnated with a 1250 ppm solution of diphenylamine controls scald but high concentrations on the wraps cause injury and off flavor. Dip treatments with Santoquin also gives promising control of scald but requires higher concentrations than diphenylamine.

Other Disorders Caused by Volatile Emanations:

Kidd and West (84) concluded that ethylene may cause lenticel spotting in certain varieties of apples. They were able to produce it by exposing apples held at 40°F to ethylene in concentrations of 1 part to 500 or to volatiles given off by ripe apples. Baker and Maxie (5) were able to control a spotting of Rome Beauty apples by use of oiled paper wraps and by air purification with activated carbon. This furnished the evidence that it is caused by apple volatiles capable of being removed by oiled wraps or activated carbon and therefore, the volatile is not ethylene. Hartman (52) reported that Anjou pears develop a superficial type of scald more like apple scald than the usual form of pears scald which extends deeply into the flesh and has a foul odor.

TRANSPIRATION

Transpiration is the loss of water from living tissues in the vapor form. The leaves are the main transpiring organs, but the fruits transpire also. After fruits are harvested they continue to transpire. After harvest they have no means of obtaining water so that transpiration losses cannot be compensated for by gains in water from any source. Hence transpiration after harvest can only be recorded as a "net loss" to the fruit.

Fruits should reach the consumer while they are still crisp and juicy. Most of the fruits are 85 percent or more water, and even a comparatively small loss in total water content means as appreciable reduction in eating quality. It has been found that storing fruits at relative humidities below 85 percent

is likely to result in wilting or shriveling of the skin after prolonged storage. With certain varieties which are not supplied with enough natural wax, relative humidities higher than 85 percent may be required ( ).

Factors Affecting the Rate of Transpiration:

1. Water vapor pressure deficit:

All liquids have a certain vapor pressure. The water gradually evaporates if the moisture content of the air is not too high. If the air is not saturated with moisture, the air will have a low vapor pressure and water will evaporate rapidly. This is true because gases always move from a point of high to a point of low concentration. As long as there will be a difference between the water vapor concentration of the air and that of the air at the surface of the water, there will continue to be evaporation. The difference in the vapor pressure of the water surface and that of the surrounding air is called "vapor pressure deficit." Temperature and relative humidity can affect the movement of water vapor.

The relative humidity may be defined as the percentage of saturation of the atmosphere with water vapor at any one temperature. Warm air can hold more water in the vapor form than cold air can. It is possible to have a relative humidity of 100 percent in both the atmosphere of a storage room and in the intercellular spaces of the fruit and yet have a vapor pressure gradient.

Smock (132) stated that the fundamental reason why fruits transpire is that there is a difference between the water vapor pressure of the fruit's internal atmosphere and that of the surrounding atmosphere. The relative humidity of the internal atmosphere of apples is presumed to be 100 percent

under normal circumstances. Hence, if the apples are held in an atmosphere with a relative humidity of less than 100 percent, water vapor moves out of the fruit into the atmosphere.

He also suggested that the relative humidity is not the only atmospheric factor that influences the rate of transpiration of fruits. Transpiration may be more rapid in a given lot of apples at 85 percent relative humidity at 36°F than in a similar humidity at 32°F. This is due to the temperature factor as it affects vapor pressures. As the temperature is raised from 32°F to 36°F the vapor pressure of water rises and the difference or deficit between fruit and atmospheric vapor pressure is increased and thus increasing the transpiration rate.

Another interrelation of temperature and vapor pressure is found with storage of warm fruit in cold storage. Transpiration is relatively rapid until the fruit temperature reaches the air temperature. As assumed if apples at 73°F are moved into a cold storage room at 32°F with 100 percent relative humidity, the water vapor pressure of the interval atmosphere of apples will be 23.8 mm. The water vapor pressure of the atmosphere of the cold storage room will be 4.6 mm of mercury only. This deficit will be about eight times as great as the theoretical deficit occurring in the situation where apples, with an internal atmosphere of 100 percent relative humidity, are held in air at 32°F with a relative humidity of 50 percent. The most striking illustration of this effect of temperature difference, as it affects vapor pressure deficit, is seen in case of a rapidly transpiring plant product, lettuce, moved into a storage and sometimes showing wilting under these conditions. Apples do not transpire rapidly enough to show

this wilting, but certainly the effect on transpiration is measurable. The only apparent practical solution to this problem is to accomplish rapid cooling after fruits are placed in storage. It has also been found that transpiration of apples is directly proportional to the water vapor deficit in any situation. Though most of the studies have been conducted with apples, the general effects of vapor pressure differences seem to apply to other fruits, too.

Curtis (27) stated that there is another temperature effect on transpiration that may affect the locus of transpiration. It has been found that if one side of an apple is colder than the other in storage, there will be a distillation of water from the warm side to the cold side, leaving the warm side withered. This phenomenon may be explained by the fact that on the warm side of the fruit the vapor pressure is greater than on the cold side and the vapor passes to the low pressure side of the fruit through the intercellular spaces interlacing from one side of the fruit to the other.

2. Effect of time of harvest:

Smock (132) found that apples and pears picked in a rather immature condition shrivel faster in storage than fruits picked at the proper time. It has been seen that Golden Delicious apples harvested September 1st, showed more wilting than fruits harvested October 1st. Thus it has been assumed that early picked fruits transpire faster in storage than more mature ones. However, Pieniazek (120) found that late picked fruits may actually transpire at a faster rate than earlier picked fruits in storage.

According to Smock (132) the respiration rate of apples and pears varies with the age of the fruit, but no correlation has been found between

respiration and transpiration. As fruits grow old the rate of transpiration eventually diminishes after the postharvest peak has been reached. This is probably due to differences in the physical structure of the skin and to internal factors.

3. Effect of fruit size:

Transpiration is a surface phenomenon. Thus a bushel of small apples and pears will transpire at a faster rate than a bushel of large apples and pears. In other words, a bushel of small apples and pears will lose more weight in storage than a bushel of large apples and pears. Pieniazek (120) found that transpiration rate of several varieties of apples in storage is directly proportional to the surface area of the fruit. Because of this strong relationship between fruit surface area and transpiration rate, he suggested that transpiration losses should always be expressed on a surface area basis rather than on a fruit weight basis as they usually are.

4. Physical Nature of Fruit Skin:

Smock (132) found that the transpiration from apples and pears is unlike evaporation from a free surface of water surface. The skin of the fruit acts as a natural deterrent to the passage of water vapor out of the fruit. Just how much the skin of the fruits of different varieties and the skin of fruit of the same variety affect transpiration rate is not entirely clear.

Cummings and Lombard (24) stated that the thickness of the skin and of the cuticle, in particular, markedly affects the rate of transpiration. It was found that Golden Delicious apples shrivel badly in storage, as it has a thin cuticle, and that the skin inhibits the transpiration is

inescapable. Smith (129) found that a unit area of free water surface will evaporate 70 times more water than a unit area of uninjured fruit of an apple per unit of time. Marshall (100) reported that when the waxy layer on the surface of the fruit is removed by brushing or by washing with severe treatments, the rate of withering is greatly increased.

White it is quite true that the nature of the skin or the cuticle in particular may have some bearing on transpiration rate, Pieniazek (120) found no good correlation between cuticle thickness and the rate of transpiration. Smock (132) stated that a more likely reason as to why Golden Delicious apples shrivel badly is the fact that there are numerous breaks and cracks in the cuticle. The cuticle on this variety is not continuous as it is on most varieties and the cracks leave the expidermis exposed and uncovered.

Pieniazek (120) also reported that other skin factors that may affect transpiration rate are the number and size of lenticels. He found that about 70 percent of apple transpiration is cuticular and about 30 percent enticular. Cummings and Lombard (24) stated that cell wall thickness and arrangement of cells in skin may also be factors in determining transpiration differences between various varieties.

5. Effect of Air Movement:

Smock (132) stated that transpiration would be accelerated by air currents passing over fruit in storage if a vapor pressure gradient exists. The magnitude of this effect is of commercial interest because some storages operate with forced air circulation, others only with slow natural convection currents.

Smith (130) reported that accelerated transpiration rates of 30 percent to 100 percent are found when air currents of varying velocities are used with apples. Some objections have been raised against this experiment as it is difficult to attribute differences in transpiration to velocity rates alone in studies where humidity values are not specified. However, this objection has been overcome by moving apples through still air at a known velocity by means of a turntable.

Pieniazek (120) found that the effect of air movement or transpiration is negligible, not exceeding an increase of 5 percent. The effect is very small when high relative humidities are used. He further stated that for apples, at least, high velocities of air do not markedly affect transpiration if the humidity is as high as it should be. The advantages of rapid cooling with moving air would doubtless outweigh the disadvantages of slightly increased transpiration.

THE METABOLISM OF POSTHARVEST FRUITS

A. Oxidations and Respiration

1. Mechanism of Oxidations:

Oxidation is carried out through the series of reactions known as the "Krebs" or "Tricarboxylic acid" cycle; based on the evidence tending to confirm the Embden-Meyerhof-Parnas theory of the course of glycolysis from sugar to pyruvic acid. Evidence has also been produced indicating the possibility of an alternative and more direct pathway in the breakdown of sugar in respiration, in the course of which pentose and heptose sugars are

produced, and to which various names have been given - "direct oxidation pathway," "pentose-phosphate pathway" and "pentose shunt."

Though the last decade has been a period of very considerable activity in the field of plant respiration, the greatest and most spectacular advances in the subject have concerned the path of degradation of the substrate into carbon dioxide and water, and the chemical mechanisms by which this is brought about. A respiratory substrate is a substance which is degraded in a plant with release of energy which is thus made available for the maintenance and growth of the plant. In normal aerobic respiration this degradation is affected by oxidation, and it has long been recognized that carbohydrates and fats serve as respiratory substrates with the end products carbon dioxide and water. However, plant acids and, in starved cells particularly, proteins might be used as respiratory substrates, too. Furthermore, it is to be expected that intermediate products in the breakdown of carbohydrate or other material to carbon dioxide and water or other products should be utilized as respiratory substrate, and it has been found by various investigators that various supposed intermediates are utilized in this way.

Fidler (31) working with apples found that the loss of carbohydrate and acid accounts quantitatively for the production of carbon dioxide and alcohol, both in air or in nitrogen. The presence or absence of oxygen is without effect on the rate of loss of acid. And under anaerobic conditions the amount of carbon dioxide and alcohol is equivalent to the sum of the carbon dioxide and alcohol which could have resulted from fermentation of the carbohydrate lost, plus the amount of carbon dioxide which could be produced from complete oxidation of the acid lost.

2. Oxidation - Reduction Potential:

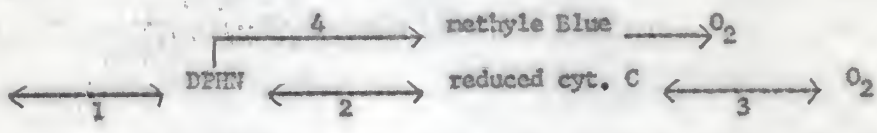
Oxidation reduction potentials in fruit extract have been investigated by several workers. Blake and Shirley (10) worked on different varieties of apples to evaluate their capacity to reduce the ceric ion and oxidation reduction potentials. They suggested that the reducing capacity of the apples is greatest during growth, intermediate during early storage, and least during late storage. Three periods of change occur in reducing capacity throughout growth and storage. No direct relation is obtained between the ceric ion reducing capacity of the apples and the concentrations of reducing sugars present. The difference in potentials (E.M.F.) between the growing and storage periods is significant at the 0.05 level (α-level); thus such evaluations should be a measure of the oxidative state of the fruit. Ulrich (152) reported that in case of pears potential is on the reduction side during the ripening period; while during overripeness, it is more on the oxidation side.

3. Respiratory Enezymes:

Biale et al. (9) reported that the respiratory machinery of the fruit, as of other plant tissues which have been studies in detail, appears to be mediated by a particulate enzyme complex, a complex located in mitochondria and competent to oxidise the acids of the Kreb's cycle, including pyruvate. The respiratory particles or mitochondria prepared from climacteric fruits are fully as active as from preclimacteric fruits. With all of the plant mitochondria, as well as with the animal mitochondria, oxidation of the substrate is linked to the uptake of inorganic phosphate and incorporation of this material into adenosine triphosphate (ATP). Rate of mitochondrial

oxidation is, therefore, limited by the availability of phosphate acceptor, such as adenylate. Inorganic phosphate is transformed in the course of complete phosphorylation into the phosphate of ATP.

According to Cheng et al. (19) the activity of the apple mitochondria is limited to succinate oxidation, while that of avocado is shown to oxidise several acids of the Kreb's cycle and to carry on phosphorylation. Mitochondria in avocado show high oxidative activity, particularly towards succinate and ⅄-Ketoglutarate. Adenylate is required for ⅄-Ketoglutarate oxidation. Thus it also appears from this that mitochondria are an organized enzyme complex. They also contain cytochrome C and are able to oxidise reduced DPN and to reduce and oxidise cytochrome C. Thus it is suggested that a normal electron transport pathway operates in the integrated enzyme complex of the cytoplasmic particles. The sequence of reactions is as follows:



Reaction 4 is shown that methylene blue can be used as an anerobic electron acceptor for the oxidation of DPNH dependent formic acid by mitochondria.

Watts and Griswold (155) reported the presence of several dehydrogenases in the pineapple fruit. Ulrich (152) reported that in apples the dehydrogenase system shows a maximum activity at about 37°C to 40°C. Tests with methylene blue show that the dehydrogenase activity drops with ripening and aging of the fruit. In green apples the malic dehydrogenase is the most active. With ripening the activity of ethanol and malic acid dehydrogenases declines. In storage the process continues further. Succinic dehydrogenase shows a rise in activity up to harvesting, then shows a rapid decline. The various kinds

of apples show considerable differences in dehydrogenase activities.

4. Terminal Oxidases:

James (72) found that in different tissues, even in the same tissues, more than one oxidase may be involved in the final transfer of hydrogen to molecular oxygen during operation of the Kreb's cycle. Evidence for the action of a particular oxidase rests partly on a demonstration of its presence in the tissue concerned, partly on the observation of the effect adding a substrate of the oxidase to the medium containing the tissue, but more especially on the effect of inhibitors of the various oxidases on the respiratory activity of the tissues. Thus, of the three oxidases supposed to be mainly responsible for the terminal oxidase action, catechol (polyphenol) oxidase is inhibited by cyanides, sulphides, azides and carbon monoxide, the last being unaffected by light, cytochrome oxidase is inhibited by the same substances, but the inhibition produced by carbon monoxide is reversible in light; while ascorbic acid oxidase is inhibited by cyanide and by diethyldithiocarbamate but not by carbon monoxide. Cytochrome oxidase, catechol or polyphenol oxidase and ascorbic acid oxidase have been recognized in a number of plants.

Ulrich (152) reported that respiratory activity of young lemon fruit is inhibited by cyanide, but not in old fruit. Thus, the cytochrome system is only active in the case of young fruit. At low temperatures, apple respiration is carried on largely by polyphenol or catechol oxidase. Cytochrome oxidase activity is higher in the fruit pulp than in the skin.

According to Joslyn et al. (79) there are number of instances where it has appeared that cytochrome oxidase acts as terminal oxidase, though not

necessarily the only one. However, James and Beevers (73) suggested that cytochrome oxidase, catechol oxidase and ascorbic acid oxidase are not the only one of the terminal oxidases in plants. The part of the respiratory activity catalysed by metal enzymes decreases during ripening while the activity of the flavine enzymes increases at the same time. Polyphenol oxidase seems to be responsible for 50 percent of the total respiratory activity (152). Webster (156) found that the respiration of apple slices in 95 percent carbon monoxide is inhibited in the same manner in light and darkness, thus indicating the absence of cytochrome oxidase. Cheng and Biale (19) suggested that there are other terminal oxidases besides cytochrome oxidase such as ascorbic acid oxidase and polyphenolic oxidase. Ulrich (152) reported that at the last stage of ripeness lycopine could operate as a substitute of oxidase for carrying oxygen.

Thus, Watts et al. (155) suggested that not one enzyme alone is responsible for the removal of hygrogen by molecular oxygen in the respiration process, but the recent works with mitochondria shows that the cytochrome system provides the main terminal oxidase while other oxidases play at most a minor part.

5. Oxidases and Fruit Browning:

Many fruits undergo rapid changes in color following mechanical or physiological injury during harvesting and storage. Such color damage in fruit products is accentuated during preparation for processing by canning, dehydration or freezing, and continues during freezing storage and subsequent defrosting of frozen fruits. The nature color of the product may be destroyed or marked by the formation of dark brown or reddish pigments which cause

the product to become unattractive in color. Undesirable changes in flavor, odor and nutritive value usually accompany this browning. Marked decreases occur in ascorbic acid content (or even its complete loss) as well as decreases in other oxidizable nutrients, such as carotene (161).

Enzyme-catalyzed oxidative browning has long been recognized (79). Several theories have been proposed for the nature and course of enzymic browning differing considerably in nomenclature as well as in mechanism. Another theory has been proposed that plant tissues which darken an injury contain a substance termed "oxygenase" which in the precence of air undergoes auto oxidation, yielding a peroxide. This peroxide, activated by the enzyme peroxidase present in the most plants, then brings about the oxidation of the natural phenolic substances.

Onslow (117) systematically investigated the oxidizing enzymes present in higher plants and put them into two groups - those which contain oxygenase and catechol compound; and those in which oxygenase and catechol compounds are absent, the peroxidase plants. The first group of plants discolor rapidly on injury and include the fruits of apple, apricot, banana, cherry, fig, grape, peach, pear and strawberry. The second group of plants which do not discolor on injury include citrus fruits - lemon, orange, lime and grapefruit; red currants, melon, pineapple and tomato.

Graubard et al. (37) suggested that there are three groups of phenolases - tryosinase (monophenol oxidase), catecolase (polyphenol oxidase) and laccase. The phenolases calatyse the oxidation of a phenolic substrate by molecular oxygen to some intermediary product, usually a quinone, which can then oxidase other constituents such as ascorbic acid, or other phenols. Cytochrome oxidase

in the presence of cytochrome is also known to reach with certain phenols in the presence of oxygen and to convert them into pigmented compounds similar to those observed in the phenolase.

Hackney (43) reported that the protoplasm, even in actively respiring cells, is under reducing conditions because the cellular oxidation-reduction potential is low enough to prevent the accumulation of the oxidized phenols, even if the phenol oxidase acts as a respiratory enzyme, which is not generally true. The lack of coloration in intact cells must be due either to reduction of oxidized phenols at a rate equal to that of their oxidation or to the fact that the phenolase does not act as a terminal respiratory enzyme in "oxidase" plants. In damaged tissues, discoloration appears at once showing that the phenol is either oxidized faster or is reduced more slowly than in the intact plant tissue. In the living cells the phenols may not be able to react because of their location in vacuoles, while the oxidases are situated in the protoplasm. According to James (72) the oxidase is strongly linked with the solid particles of apple and pear pulp, the enzyme is concentrated in the core and below the skin.

Ulrich (152) reviewed the postharvest physiology of fruits and suggested that orthodiphenolase, peroxidase, catalase and dehydrase are present in apple juices. In presence of oxygen the phenol is oxidized by oxidase into quinone, the latter probably being reduced to phenol by ascorbic acid by the dehydrase. When all the ascorbic acid is oxidized, the phenols can not reappear from quinones, and the juice becomes brown. However, Ingraham (71) stated that catechol oxidation by polyphenoloxidase in air is not inhibited by ascorbic acid, but the dehydrase is lacking here.

Graubard (37) stated that the enzyme system mainly responsible for browning is polyphenol oxidase, but little information is available as to the contribution of the individual substrates which are subject to darkening by polyphenol oxidase, although it has been recognized that they are, in the main, compounds containing o-dehydroxy (phenolic) groups. Chlorogenic acid, which contains such a configuration and has been found in apples and pears, does in fact take part in the enzymic browning of these fruits. As browning proceeds, chlorogenic acid decreases and three fluorescent compounds are formed from it during the reaction. Ascorbic acid and other naturally occurring compounds containing -SH groups, such as cystine and glutathione, act as inhibitors. Ascorbic acid is supposed to act by reducing the initial oxidation products of the substrates and so preventing their conversion into colored products, where as -SH group compounds inhibit the action of the enzyme itself. Seigelman (134) while working for the detection for the substrates of polyphenol oxidase, found l-epicatechol in skin extracts of apples and pears and small amounts of d-catechol in Bartlett pears.

6. Kreb's Cycle and Pentose Cycle:

As mentioned earlier, the chemical changes that take place in the detached fruit are directly or indirectly related to the oxidative and fermentative activities, collectively referred to as biological oxidations. Respiration, a process concerned with the oxidation of predominantly organic substances by the cell or by enzymatic systems derived from the cell, is restricted to the reaction requiring oxygen, whereas fermentation or glycolysis is characteristic of biological oxidations in an oxygen-free environment. Even then, fermentation may also take place in an atmosphere

containing oxygen. A distinction is made, therefore, between aerobic and anaerobic glycolysis on the basis of the conditions to which the fermenting material is exposed. The following reactions illustrate the two processes:

(1) Fermentation: $C_6H_{12}O_6$ ——— $2CH_3.CO.COOH + 4H$

(2) Respiration: $CH_3.CO.COOH + 2\frac{1}{2}\ O_2$ ——— $3CO_2 + 2H_2O$
(Pyruvic acid)

The breakdown of sugar to pyruvic acid consists of the phosphorylation of glucose or fructose to a hexose diphosphate, the splitting of the six-carbon sugar phosphate into two triose phosphate units, isomerization, oxydation of phosphoglyceraldehyde to phosphoglyceric acid, and successive transformation of the latter into pyruvic acid. The coupled oxidation - reduction results in the formation of adenosine triphosphate (ATP) from adenosine diphosphate and inorganic phosphate. ATP is also formed when phosphoenolpyruvase is changed to pyruvate. In reality pyruvic acid is not the end product of glycolysis but is converted by fermentative processes into alcohol, lactic acid, propionic acid etc.

In respiration pyruvic acid, the cleavage product of glycolysis, is completely oxidized to CO_2 and water. The metabolic pathway responsible for this oxidation is known as the Krebs' cycle. The essential features of this cycle are the condensation of a two-carbon fragment derived from pyruvate with a four carbon acid, oxalacetate, to form citrate, and the successive transformation of the latter into aconitate, isocetrate, oxal-succinate, ℓ-ketoglutarate, succinate, fumarate, malate and oxalacetate.

There are five sites in the Krebs' cycle for the transfer of a pair of electrons from substrate to oxygen via flavins, cytochromes, and cytochrome

oxidase. DPN^+ (oxidised diphosphopyrictine nucleotide) participates in three of these sites, and TPN+ in one, but pyridine nucleotides are not involved in the fifth (succinate oxidation). Whenever, an electron is transferred through DPN and TPN, a minimum of three ATP molecules are formed for each molecule of oxygen reduced. Succinate oxidation results in the generation of two ATP molecules per atom of oxygen. In addition, the free energy change when α-ketoglutarate is converted to succinate anearobically, permits the formation of one ATP molecule for each keto acid molecule disappearing. The net theoretical result is that at least 15 molecules of ATP are generated for each molecule of pyruvate oxidized in the Krebs' cycle. Thus, the energy that becomes available is much greater in respiration than fermentation.

During the last few years a number of investigations have indicated that hexose may be broken down in plants to carbon dioxide and water by a course different from that of the EMP pathway and the Krebs' cycle. This pathyway, "pentose shunt" or "pentose phosphate pathway" or "direct oxidation pathway" or "oxidative glycotysis pathway" or "hexomonophosphate shunt," involves a cycle of reactions in which phosphorylated hexose is degraded to phosphorylated pentose with release of carbon dioxide and water, and ultimate reformation of hexose from the pentose through a series of reactions in which phosphorylated sugars are the intermediates. The supposed reactions in the cycle are:

(1) Glucose is first phosphorylated to glucose -6- phosphate (enzyme hexokinase).

(2) The glucose -6- phosphate is oxidized to 6- phosphogluconate

(coenzyme 2(TPN) and dehydrogenase).

(3) By the action of the enzyme 6-phosphogluconate dehydrogenase with TPN the phosphorylated keto-pentose sugar ribulose -5- phosphate is formed and a molecule of CO_2 is released for every molecule of sugar involved.

(4) The ribulose -5- phosphate is now transformed to its aldopentose isomer ribose -5- phosphate through the action of the enzymes phoribose isomerase.

(5) Two molecules of the ribose -5- phosphate under the action of the enzyme transketolase now give rise to a molecule of the heptose sugar sedoheptulose and a molecule of the triose glyceric aldehyde, both as phosphate esters.

(6) By the action of the enzyme transaldolase a three carbon atom chain from the sedoheptulose is now linked to the carbon chain of the glyceric aldehyde with the result that hexosephosphate is formed. A second molecule of hexosephosphate is formed by the linkage of the remaining chain of four carbon atoms from the sedoheptulose with a two-carbon portion of a pentose molecule. This leaves a three-carbon portion, a triose of the pentose molecule, and two of these portions link up to produce further hexose. The enzymes involved in these two reactions are, respectively, transketolase and aldolase.

Thus for every six molecules of hexose entering into this series of reactions, five are reformed, and from the one lost six molecules of CO_2 are provided. There also arise 12 molecules of reduced coenzyme, and, if

these are oxidized by six molecules of oxygen by means of terminal oxidases there will be a net gain of six molecules of water. The overall equation for the direct "oxidation pathway" is thus the familiar $C_6H_{12}O_6 + 6O_2 \longrightarrow 6O_2 + 6H_2O$.

Ulrich (152) in his review stated that experiments on tissue slices in the Warburg apparatus show the stimulating effect on respiration of some organic acids - such as malic acid. The very low efficiency of citric acid in such experiments led to the hypothesis that the part of the Krebs' cycle involving this acid is slow or lacking in fruits. Experiments on the effects of banana extracts on hexose-phosphate and ribose -5- phosphate and on the action on respiration of sodium fluoride indicate that the pentose cycle may be active in the respiration of preclimacteric bananas, but near the climacteric rise, respiration via the EMP pathway seems to increase.

SUGARS

B.

1. Metabolism of Polysaccharides:

Hulme (68) reported that the residue after extraction of apple and pear tissue with 70-90 percent ethanol, the alcohol-insoluble residue, comprises all the polysaccharide material of the fruits together with a very small amount of "protein." The polysaccharide material consists of pectic substances, starch, hemicelluloses, cellulose, and in case of pears, lignin in the stone cells. Starch is most obviously related to changes in sugar, but evidence has been accumulating that the polysaccharides associated with

the cell wall may be continuously involved in the respiratory process of fruits, presumably, after prior degradation of sugars. A small amount of iodine-coloring starch degradation products (dextrins) in apples, at certain stages of development has also been reported by Griffiths et al. (39).

Pectic substances will be discussed in a separate section. It will be convenient, therefore, to discuss starch alone in the present section.

Starch itself is a mixture of straight-chained amylase component and a branched-chained amylopectic fraction. Amylase gives an intensely blue colored complex with iodine while the color of the amylopectin-iodine compound is blue-violet; and that their amount varies according to the varieties.

It has been found that very little starch is present when apples and pears are normally harvested and stored. Kidd and co-workers (86) stated that after a short period during which starch synthesis, as a carry-over from conditions in the fruit on trees, acts as a delaying factor, degradation of starch alone proceeds in detached apples. The rate of degradation is proportional to the surface area of the starch grains. The temperature coefficient of the degradation process is small; synthesis is more retarded at low temperature than in the degradation process.

Kulne (86) reported that in living, healthy tissue the amount of any substances present in the tissue is a result of an equilibrium between synthesis and breakdown. Based on this, as starch disappears from the stored apple it would be assumed that the equilibrium synthesis and breakdown is rapidly shifting in the direction of breakdown, and according to Kidd (86), after a certain adjustment phase, breakdown takes over completely. Starch

unlike protein, cellulose, cellwall pectin etc., is not an essential to a fruit cell. Its main function can, apparently, be taken over by its constituent parts, the sugars.

Hulme (63) was unable to find any data on the pattern of starch change in pears during growth on the tree, not indeed, during storage. The reason for this is that little starch remains in pears when harvested commercially.

However, he reported that the enzyme responsible for the hydrolysis of starch in fruits is amylase. Little data is available on the amylase content of apples and pears. Starch is not synthesized, and probably not hydrolysed either, in most metabolizing cells by amylases but by processes involving phosphorylation. Amylases are of course concerned with digestion in animals and the mobilization of starch during the germination of seeds. Until pure amylases, which hydrolyze starch in a manner similar to those of germinating barley, have been isolated from apples and pears, any demonstration of "starch splitting activity" by fruit tissue or crude preparation, therefrom, cannot be regarded as necessarily due to amylase action. Ulrich (152) in his review stated that the effect of α-amylase becomes greater at low temperature, although starch is lacking. There is a certain parallel between the rise in α-amylase activity and the possibility of ripening after cold storage; if the rise in α-amylase activity is too great, ripening is made impossible. No important changes occur in the quantities of α-amylase during the storage period.

Hulme (68) stated that pectin, hemicellulose, and α-cellulose are the resultants of certain sets of operations rather than chemical individuals. Each of these fractions is a mixture of polysaccharides . . . and the same

polyraccharides may be present in more than one fraction. The study, on changes in the cell wall during the ripening of the pear in terms of the polysaccharides corresponding to individual sugar residue, shows that the alcohol-insoluble residue contains the following polysaccharides: - pectin (flucosan, galactan, xylan, araban, and polygalacturonic acid); hemicellulose A (glucosan, galactan, xylan, araban, and uronic acid); hemicellulose B (as A, but minus araban and uronic acid); d - cellulose (glucosan, galactan, mannan, and xylan). Changes in these various fractions have been measured (in terms of sugars produced in hydrolysis) during storage of "conference pears" at 10° and 15°C. The greatest change occurs in the xylans and arabans. Both these types show a rapid rise during a change in the texture of the fruit from "sleepy" to "slushy", and polygalacturonic acid also rises over this period. Cellulose shows a steady fall in storage.

Chanda et al. (18) suggested that xylan is a branched chain structure which appears to have 115 D-xylopyranose units in a chain with a single branch but carrying in addition a terminal D-glucopyruronic acid residue at one point.

Changes in the hemicellulose content of Bramley's Seedling and Worcester Pearmain apples during development on the tree and in storage have been studied by Widdowson (159), on the pear by Jermyn and Isherwood (75), and on McIntosh apples by Krotkov and Nelson (91). Unlike pears there are not lignified (stone) cells in apples so that lignin is not present in any quantity in alcohol - isoluble residue. A slight but steady fall, in hemicellulose during storage, has been found.

Hulme (68) in his review reported about the changes in cellulose content

of apples during growth and cool storage. The cellulose content rises rapidly in early July but falls again from the middle of July until just before normal harvesting time (mid-October); and reaches its highest proportion, about 22 percent in the alcohol-insoluble residues, about the end of October.

2. Metabolism of Soluble Sugars:

Sugars (including starch) which on hydrolysis in the fruit reappears as sugars form almost the entire substrate for metabolic processes yielding energy. The "energy level" of the fruit is expected to determine the length of its "life" on detachment from the tree.

The bulk of the sugar in both apples and pears is comprised of sucrose, glucose, and fructose. Most of the studies of sugar changes during growth and storage of the fruits have been carried on with changes in these sugars (glucose and fructose being often determined together as total reducing sugars). Hulme (68) in his review has referred to the variation of glucose and fructose in English varieties of apples. In the juice of apples sucrose varies between 6.6 and 56.8, glucose between 12.3 and 58.0., and fructose between 69.2 and 113.8 gms per liter depending upon the variety. The juice of pears generally contains less sugar than that of from apples. After analysis, it has been found that sucrose varies between 1 and 24, glucose between 5 and 35, and fructose between 65 and 112 grams per liter. In young fruits the proportion of the three sugars is about the same as in the mature fruits. Thus, in both apples and pears fructose is in excess of glucose, and sucrose is the least abundant of the three sugars.

The existance of xylose and galactose has been reported by several

workers. Xylose has been found in the juice of Japanese apples, and in the juice of Williams pears. Ash and Reynolds (2) reported the presence of galactose in pears and in trace amounts in apples. Siegelman (126) investigated the presence of sucrose, fructose and xylose in the skin of Grimes Golden apples and Bartlett pears. Though xylose is the only pentose so far detected in these fruits, ribose at least is also formed for incorporation in the nucleic acids universally present in living tissue.

Ash and Reynolds (2) have also detected two ketooligosaccharides in several varieties of pears in small amounts at least. On hydrolysis, one of these oligosaccharides gives chromatograms on which xylose, glucose and fructose could be detected. Their work suggests that transfructosidation can proceed in fruits as in other plant tissues.

Tutin (151) detected sorbitol, the hexahydroxyl alcohol corresponding to sorbose, the ketose, from apples, Strain (145) and Martin (101) from pears. Kidd et al. (85) detected considerable amounts of sorbitol from Conference pears and reported on the changes in the fruit, during storage at 10°C. He suggested that sorbitol is transferred into fructose in these fruits. Ash and Reynolds (2) isolated hexitol and also a cyclitol, which they considered as probably mesoinostil, from apples and pears. The amount of hexitol present is about the same as that of sucrose.

Axelrod and Seegmiller (4) found a formation of radioactive sucrose (after infiltration of radioactive glucose into apple discs), but not of free fructose. This process is inhibited by exclusion of oxygen.

Very many papers, on the course of the change in sugars in fruits at various stages of development on the tree and during the storage, have been

reported. Fructose is the most prominent sugar except during the first few weeks of development. Sugars increase steadily up to, and indeed rather beyond, the time at which the fruit is harvested commercially, fructose being in considerable excess at the end of growth. Works, reported by Archbold (1) and Kidd et al. (88) on the changes undergone in the sugars when apples and pears are placed in storage at different temperatures, show that fructose is the most abundant sugar.

Hulme (60) worked on changes in sugars in Bramley's seedling apples picked at different stages of growth and stored at 12°C. It was found that starch synthesis and hydrolysis are not directly related to the increase or decrease in reducing sugars. Reducing sugars continue to increase long after starch has disappeared in fruits picked toward the end of the season. Changes in starch and in sucrose are generally considered as being linked processes, although there is far more sucrose synthesized than could be accounted for by starch lost. Kidd (86) suggested that no starch synthesis, only degradation, occurs on removal of fruit from the tree (the starch, where present, does not present an equilibrium between synthesis and hydrolysis).

The rise in sugars at one period or another in postharvest fruits is also considered in the work of Krotkov and Nelson (91) who suggested that the bulk of these sugars come from some (85 percent) alcohol-soluble substances which are precipitated from the extracts during clearing with lead. Thus, it has been suggested that a component of the "organic acid fraction" is involved and, indeed, Krotkov et al. (92) concluded that the carbohydrate and acid metabolisms of the apples are closely related, but that the relation is not a simple one.

Smock and Neubert (138) quoted the work done by Magness et al. (98) who measured the changes in total and reducing sugars in apples stored at different temperatures. Lowering of the temperature resulted in a retardation in the loss of both reducing sugars and sucrose. Griffiths et al. (42) worked on the changes in glucose, fructose and sucrose in mature Bramley's seedling apples in storage at various temperatures. He found that there is a small loss in weight during storage, and these weight losses do not account to any appreciable extent for the increase in glucose and fructose during the storage period, even at the higher temperatures where weight losses are greatest. Starch falls to negligible proportions in the first 50 days at 1°C and in less than 20 days at 15°C. Changes in glucose and fructose appear to be least affected by temperature, the most striking changes being in sucrose. Although the increase in sucrose may be partly a result of hydrolysis of starch, the increase is greater and lasts longer than can be accounted for in terms of loss of starch.

Kieser and Pollard (90) worked on the juices of several apple varieties, and found that sucrose changes considerably more than reducing sugars during storage of the fruit at 3-5°C. Hulme (68) in his review reported that durthe ripening of pears at 15 C, sucrose increases rapidly and then falls as rapidly, while reducing sugars undergo much less change.

Onslow et al. (110) analysing the sugar changes in Worcester Pearmain and Bramley's Seedling apples concluded that glucose is constantly being converted to fructose in the fruit, making "spot" determinations of the two sugars misleading; and the fructose formed is immediately condensed to sucrose. This lead to the hypothesis that sucrose at the point of inversion

is the main substrate for respiration in the form of the y-fructose liberated. Onslow et al. (117) stated that sucrose never falls to zero but that there is a critical residual amount, the basal sucrose content, which never enters into the respiration processes. They associate a high level of this basal sucrose content with bad keeping. But Archbold (1) associated a high level of basal sucrose merely with late harvesting. Onslow et al. (118) stated that the susceptibility of apples to tissue breakdown at low temperatures is associated either with a failure of the cell mechanism to convert glucose to fructose or with the absence of any mechanism in the cell for the flycolysis and respiration of glucose. The level of sucrose is certainly very susceptible to storage conditions, and it is therefore possible that the level of sucrose, or its rate of change in storage, might be a major factor in determining the storage life of the fruit.

And finally Kidd et al. (86) suggested that in stored apples from the time of the disappearance of starch, the change in sucrose fits a curve with the following formula: log (c-m) + b - at, in which C is the observed sucrose content, m the steady state value, b the initial value, a the rate constant, and the final low level (steady state) value may be higher for Worcester Pearmain than for Bramley's Seedling apples.

ORGANIC ACIDS

C.

1. Acids found in fruits:

After the new chromatographic methods came into use, numerous papers

have been published dealing with the distribution and characterization of organic acids in fruits. Since 1951 rapid strides have been made, as the result of the use of paper chromatographic techniques in the detection of organic acids in apples and pears. The following table gives a list of the acids which, in addition to malic and citric acid, are now known with some certainty to be present in these fruits.

ORGANIC ACIDS KNOWN TO BE PRESENT IN APPLE AND PEARS

APPLE			PEARS		
Whole fruit or juice of whole fruit	Pulp	Peel	Whole fruit or juice of whole fruit	Pulp	Peel
quinic	quinic	quinic	quinic	quinic	quinic
glycolic	shikmic	shikmic	glycolic	shikmic	shikmic
succinic	succinic	citramalic	succinic	glycolic	glyceric
lactic	glyceric	glycerlic	lactic	mucic	citramatic
galacturonic	-ketoglutaric	-ketoglutaric	galacturonic		
citramalic	pyruvic	pyruvic			
	oxalacetic	oxalacetic			
	glycoxylic				
	isocitric				

Evidence discussed by Nitsch ([114]) shows that both acids and sugars enter the fruit are preformed. However, Tomkins (148) concluded that at best a part of the acids are formed in the fruit from carbohydrate. Krolkov et al. (91) found a close relationship between carbohydrate and acid metabolism in fruit

attached to the tree but concluded that the relation is not a simple one. Possibly the acids found in the fruit arise from both sources. There is not critical evidence in this point, but it is perhaps significant that an appreciable increase in the titratable acidity has never been observed when once the fruit has been detached from the tree. Some small increase in titratable acidity may occur in the first day or two after picking in immature fruits, but this may be due to the minor acids such as quinic acid.

Hulme (68) stated that in the light of present knowledge of the position of acids in plant metabolism, it appears probable that the acids present in small amount may play a part of equal or even of greater importance than the major acids, in the general metabolism of apple and pear fruits. These major acids may indeed be reservoirs for feeding acids into a complicated cycle of acid transformations (the cycle itself providing energy, through energy-rich phosphate bonds, much greater than that provided by the direct oxidation of the acids), or they may be "sinks" of acids thrown out by such a cycle. Such alicyclic acids as quinic and shikimic are almost certainly moving in metabolic pathways other than those involving the open chain acids such as malic and citric.

2. Fluctuations of the different acids:

From the early work of Haynes (54) numerous papers have been reported on the changes in the titratable acidity of apples after removal from the tree, especially after detachment at the normal time of harvest. Most of this work has been on the pulp of the fruit, and since the bulk of the acid at this stage is malic acid and the amount of cation (mainly potassium) cannot change, changes in titratable acidity may reasonably be taken as

truly representative of the gross change in malic acid. Kidd and Hanes (82) found that drifts in pH at various temperatures are explicable on the basis of decreasing concentration of free malic acid, in apples in storage, in the presence of a small amount of monobasic salt which remains constant in concentration.

Kieser and Pollard (90) working on changes in acid for a number of varieties of apples, stored at 3-5°C, found that after an initial period of a few days when the acid content remains constant or may rise slightly, there is a steady logrithanic fall at low or high temperatures. Haynes (54), Kidd et al. (87) and Fidler (31) found that the rate of loss is constant, and it is not affected by the climacteric rise in respiration and it appears to be constant for a given variety of apple.

Fidler (31) showed that titratable acid (malic acid) in apples in storage at 12°C is lost as rapidly in pure nitrogen as in air. This is most surprising since oxidation of malic acid would require large amounts of oxygen and if "oxidized" in absence of oxygen would involve the formation of large quantities of highly reduced compounds.

Very few papers have been published on changes in the titratable acidity in detached pears. Kidd et al. (85) stated that the acidity of Conference pears is very low as compared with apples. It shows no significant change during storage at 10°C until physiological breakdown of the tissue occurs; subsequently the acid content falls considerably. Leonard (95) found that acids of Bartlett pears decrease during ripening except in the later stages.

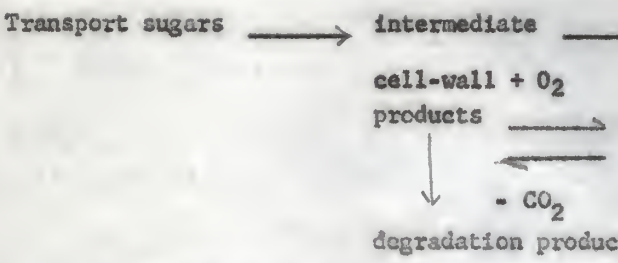
Turner (150) found that pulp of Granny Smith apples contains small

amounts of oxalic and tartanic acids. He found a very small erratic loss of malic acid from apples stored at 0°C.

Hulme (68) worked on the changes in citric, citramalic (present in peel only), quinic, and shikimic acids in Bramley's Seedling apples stored at 15°C. In the pulp citric acid rises rapidly during the first 15 days and then more slowly at the end of 100 days of storage. Shikimic acid appears in very small quantity near the end of the storage period. In the peel of the apples, citric acid content remains low and fairly constant during storage. Quinic acid follows a similar pattern to that obtaining in the pulp, with a peak at about 40 days. Shikimic acid rises steadily, with an increase in rate when quinic acid commences to fall, from 5 at commencement to 8 mg per 100 grams at 100 days. When the fruit is harvested, the peel contains no citramalic acid. After 25 days, it has been found that 10 mg per 100 grams is present and this rises by the end of the storage period (100 days) to 25 mg. Therefore, one of the most intriguing aspects of this new knowledge of the "micro acids" is the relatively large changes they undergo during the life, especially the off-the-tree life, of apples and quite large variations have been found from season to season and variety to variety. It has also been found that, in pears, this pattern is even more mobile.

3. Pathways of Synthesis and Breakdown:

"Acid" in general sense is mostly involved in "respiration." Kidd et al. (87) feel the formation of acid in the tree is due to processes requiring the presence of oxygen. They consider that it is linked with the systems involving cell-wall formation through intermediate "cell-wall products" according to the following scheme:



But there is no biochemical evidence to prove or disprove this hypothesis.

Robertson and Turner (125) suggested that organic acids are transported from the leaf in approximately the proportions in which they occur in the leaf tissue and that only as the fruit develops, and its "enzymatic capacity" increases, they are transformed gradually to those characteristic of the mature fruit.

Kidd et al. (87) reported that the loss of acid which takes place in stored fruit is due to decarboxylation which does not require the presence of oxygen. This also agrees with the work done by Fidler (31). Turner (150) suggested his results on malic and citric acid changes on the basis of the Krebs citric acid cycle, but there is no firm evidence to date for the operation of "full" Krebs cycle in the apple.

A possible role for quinic and shikimic acid is as precursors of aromatic ring compounds. An essentially active role for these acids is consistent with the considerably higher concentration in the more vigorously metabolizing peel tissue. Peel tissue is also relatively rich in phenolic substances. These acids, small in absolute amount, also fluctuate rapidly, and the appearance of shikimic acid in pulp tissue only when the fruit is "run down" may be a reflection of the disorganization and slowing down of the cellular processes

at this stage. Hulme (61) has suggested that shikimic and quinic acids might form a dehydrogenase system analogous to the succinic-fumaric malic system:

$$\text{Dihydroshikimic acid} \underset{+H}{\overset{-H}{\rightleftharpoons}} \text{Shikimic acid} \underset{-H_2O}{\overset{+H_2O}{\rightleftharpoons}} \text{quinic acid}$$

But there is no experimental evidence to support this theory.

Hulme (65) stated that citramalic acid has never been found in the pulp of apples so far. It appears in the peel only at maturity. This acid is readily oxidized in vitro to acitoacetic acid which in turn readily breaks down to acetone. Meigh (107) found that in apples stored at $4^{\circ}C$ acetone is the most abundantly produced volatile carbonyl compound and that in some varieties the amount evolved increases with storage. It seems, therefore, that at least some of this acetone might arise from the citramalic acid in the peel of the fruit. The pathways leading to the production of citramalic acid are quite unexplored.

Ulrich (152) reported that organic acids do not contribute to the increase in sugars during climacteric and are not formed during the decrease in sugars in the postclimacteric period. They are formed by carboxylation. It has been found that apples exposed for 18 hours in darkness to CO_2^{14} fixed C^{14} into malic, aspartic, and glutamic acids, 2-alanine and serine. Malic acid and amine acids contain over 80 percent of C^{14} incorporated in the involatile compounds. Carbon dioxide fixation increases as the CO_2 concentration in the atmosphere is raised. Hulme (67) stated that CO_2 injury of apples stored in an atmosphere containing 20 percent CO_2 is

accompanied by an increase in succinic acid content of the tissue. Ulrich (152) in his review stated that Williams pears, stored at $0^{\circ}C$ in air enriched with 10 percent CO_2, accumulate malic and succinic acids in the tissues.

Hulme (62) put forth his views on the biosynthesis of l-quinic acid in apples. In vitro, quinic acid may lead to citric and aconitic acids by oxidation and to protocatechuic acid and shikimic acid. By combining with the relatively insoluble caffeic acid, quinic acid gives chlorogenic acid in apples, Hulme (63). It is not certain, however, that shikimic acid comes from quinic acid in vitro. It has been suggested that quinic acid might provide a link between aliphatic and aromatic compounds in plants and that shikimic acid might be the first stage from quinic acid in the desaturation.

PECTIC SUBSTANCES

D.

1. Definition and Nomenclature:

The pectic substances are carbohydrates or, more exactly, carbohydrate derivatives. D-galacturonic acid is the main building unit of the pectic substances. On the basis of the term "polyuronide," first used by Smolenski, the pectic substances are polyuronides composed mostly of anhydrogalacturonic acid residues, although some authors (102,143) still maintain that other carbohydrates, for example arabinose, galactose, sorbose, rhamnose are attached to the chain of anhydrogalacturonic acid units.

The revised nomenclature of the pectic substances was reported to the

American Chemistry Society in 1943 and adopted as official in April, 1944.

"Pectic substances": "Pectic substances" is a group designation for these complex, colloidal carboxydrate derivatives which occur in, or are prepared from, plants and contain a large portion of anhydrogalacturonic acid units which are thought to exist in a chain-like combination. The carboxyl groups of polygalacturonic acids may be partially esterified by methyl groups and partly or completely neutralized by one or more bases, according to Kertesz (80).

Kertesz (90) also stated that the term pectic substance appear to be the most satisfactory general designation for this group of compounds. It is undesirable to use pectin or pectins for this purpose. These substances are described as carbohydrate derivatives in contrast to carbohydrates. In general, pectic substances are distinguished from polysaccharides by the possession of carboxyl groups. The carboxyl groups are part of the anhydrogalacturonic acid units characteristic of all pectic substances.

According to Kertesz (80) the followings could be defined as follows:

"Pectinic acids": "The term pectinic acids is used for colloidal polygalacuturonic acids containing more than a negligible proportion of methyl easter groups. Pectinic acids, under suitable conditions, are capable of forming gels (jellies) with sugar and acid or, if suitably low in methoxyl content, with certain metallic ions. The salts of pectinic acids are either normal or acid pectinates."

"Pectin": "The general term pectin (or pectins) designates those water soluble pectinic acids of varying methyl ester content and degree of neutralization which are capable of forming gels with sugar and acid under

suitable conditions."

"Pectic acid": "The term pectic acid is applied to pectic substances mostly composed of colloidal polygalacturonic acids and essentially free from methyl ester groups. The salts of pectic acid are either normal or acid pectases."

2. Occurrence and Distribution of Pectic Substances in Plants:

Kertesz (80) write that pectic substances occur in most, perhaps in all, plant tissues. Generally speaking, they are found in relatively large amounts in succulent, soft tissues composed chiefly of primary walls, and under conditions of rapid growth and high water content. During the process of lignification the content of pectic materials in plants usually decreases and in hard tissues such as woody they constitute only a negligible fraction of the total plant substances.

Kertesz (80) also stated that the bulk of the interior of mature cells is occupied by a single large cavity, the vacuole. This is filled with water in which a great variety of substances are dissolved or dispersed. Although the occurrence of pectic substances in the cell sap has been observed, there is doubt whether such dissolved pectic substances are common. The pectic compounds of the cell wall are formed in the cell itself and are only later deposited, which makes the cell sap the primary source of pectic substances in the plant tissue. There is no doubt, however, that some of the dissolved pectic substances may at times originate from the insoluble pectic constituents of the cell walls and the middle lamela. There is ample evidence that such dissolution takes place, especially as the tissues mature and disintegrate. As a consequence, pectic materials in the state of transition can also be

found in plant tissues. The solubility of pectic substances in the cell sap might be governed, at least partly, by the degree of methylation.

In young tissues the cell wall consists of a single layer, while in older tissues it is composed of two or more layers. A well developed cell wall is composed of the following three main layers:

1. The middle lamela or intercellular substance is formed from the cell plate during cell division and is shared by adjacent cells. This is composed largely, or entirely, of pectic substances.
2. The primary wall is composed of cellulose, hemicellulose, pectic substances and lignin.
3. The secondary wall does not contain pectic substances.

According to Kertesz (80) the pectic substances of the middle lamela are deposited in a single or double layer by the plasma membranes and undergo changes in form, quantity, and characteristics during the development of the plant. This mass is often increased by the secretion of further pectic materials from the adjoining cells into the spaces formed when they are rounding off. The primary cell wall, contrary to the middle lamela, is rich in typical protopectin. The majority of root hairs contain all inner membrane composed mostly of cellulose and an outer one consisting of pectic substances.

3. Metabolism:

Pectic substances are largely associated with the cell wall, and in consequence, the softening of fruits might be expected to be a function of pectic changes. Generally soluble pectin increases during ripening at the expenses of protopectin, but the qualitative changes of pectin are not well known. Bonner (11) quoted Haller's (45) data to show the correlation, in

apple fruit in cold storage, of decrease in protopectin content with increase in pectin content and decrease in firmness as measured by penetrometer. However, Haller et al. (46) stated that softening in storage is apparently due to the conversion of the insoluble pectic substances, principally protopectin, into soluble form. The storage temperature affects the rate of change in pectic substances, but does not affect the general trend which is a more or less rapid fall in net protopectin accompanied by a concomitant rise in net water-soluble pectin. Later there is a period in which both fractions remain constant; the fruit becomes mealy, soluble pectin decreases rapidly and protopectin again increases somewhat.

Hulme (68), in his review, gave a typical example of changes in protopectin and soluble pectin as related to changes in the hardiness of apples. He stated that at the four temperatures of storage, net fall in protopectin is in each case almost exactly balanced by increase in soluble pectin. Trends in both fractions are not all in one direction; the fluctuations are probably greater than can be accounted for by experimental error. Changes in the hardiness of the apple follow very closely the changes in protopectin content. The rise in protopectin toward the end of the storage period is not always evident in stored apples.

Hulme (68) reported that there is a relationship between the softening of apples and the total organic acid content. During storage at 4°C from December to June, the ratio remains fairly constant with a gradual rise up to March, the rise being more prolonged in some varieties than in others. The rise in this ratio appears to be almost entirely a reflection of a fall in organic acid.

Poast and Phillips (121) claimed that in apples stored at 0°C, changes in "soluble pectin" (Pectin and pectinic acids) fall into three phases. The "climacteric" peak of the first phase coincide with maximum eating quality. The high quality and longe storage life coincide with low levels of soluble pectin and that in apples receiving a high level of nitrogen the pectin climacteric is high and sharp, as high nitrogen apples are poor keepers.

Hulme (68) also reported that the situation in the pear is much more interesting and more readily followed. The change from a hard condition to the soft "melting ripe" condition so essential to eating quality in pears takes place rapidly at ripening temperatures, about 20°C. Changes in pectic substances are rapid during this softening process. A few varieties of pears never ripen on the trees. Most pears that do not ripen after detachment will not ripen (soften) normally if maintained too long at cold or moderately cold temperatures. They become sleepy and only soften with concomitant browning of the tissue when brought to higher temperatures.

Most comprehensive study of pectic changes in pears during growth on the tree and during storage and ripening has been reported by (68), in his review. Unlike the apple there is no rapid fall in total pectin during June. In fact total pectin remains constant throughout June and two thirds of July. A relatively rapid decrease in total pectin then occurs which lasts until about the 20th of August. From then until the 10th of September total pectin remains constant followed by a rapid fall to the beginning of October. The changes in total pectic substances during growth are largely due to changes in protopectin. A drop in protopectin does not result directly in an increase

of soluble pectin; it is possible that the shifting equilibrium between the "pectic fractions' is a reflection of a utilization of these substances in respiration. The respiration of pears falls rapidly from May until the end of July and low value for soluble pectin with a gradual rise during June, July, and early August lends support for this suggestion.

He further reported that pectic changes in pears maintained from the 29th of August to the end of November at 0°C are small. Where the pears are given an intermediate period of 15°C, pectic changes are appreciable, most of the change occurs during this warming up period. The most profound changes occur in fruit stored at 0°C up to the 15th of November and then kept at 15°C for 10 days. Fall in protopectin is accompanied by rise in soluble pectin though the quantities involved are not identical. In general, the decrease in hardiness of the fruit, as with apples, tends to accompany fall in protopectin.

Date and Hansen (26) reported on the different behavior of different varieties of pear. They also throw light on the development of "sleepiness" in pears. Pears harvested at the normal time were stored at -1.11 to 0.56°C. Three varieties of pears were examined: Bartlett, Bose and Anjou. In cold storage, protopectin increases to a maximum and then declines steadily to the end of the storage period. They emphasize that as Bartlett and Bose pears progress in cold storage, their ability to hydrolyse protopectin on removal to higher temperatures declines and in January, after 12 days at 20-21.11°C, they still contain more protopectin than when originally harvested. Towards the end of the storage period, the Bartlett pears failed to soften to

"melting ripe" when removed to ripening temperatures so that it would appear that "sleepiness" in pears may be associated with inactivation of protopectinase. The Anjou pears were in a preclimacteric state at harvest, and exhibited no fall in soluble pectin on ripening immediately after harvesting, and retained their ability to convert protopectin to soluble pectin and to soften on ripening to the end of the storage period.

Hulme (68) state: "To summarize, the gross overall pectic changes during normal ripening of apples and pears appear to involve firstly a hydrolysis of protopectin resulting in an increase in soluble pectin. This in later stages itself disappears, presumably through degradation of the polygalacturonic acid chains since the viscosity of the extracted pectin decreases as ripening proceeds."

4. Pectic Enzymes and their Mechanism:

No account of the pectic changes taking place in apples and pears can disregard some consideration of the mechanism responsible for these changes. It has been found that pectin, hemicellulose, and cellulose are both broken down and synthesized during the physical changes which take place during ripening. This being so, enzyme processes must be involved.

Two kinds of enzymes seem to be involved in the breakdown of pectic materials. Pectin polygalacturonase (P.G.) - the enzyme responsible for the breakdown of pectinic acid (more strictly, pectic acid) to shorter chain length polygalacturonic acids and even to galacturonic acid itself. "Pectase" or pectin methylesterase (P.E.) is responsible for demethylating pectinic acids. "Pectinase" may be regarded as a mixture of P.G. and P.E. since Jansen and MacDonnell (74) reported that P.G. has but slight action on a methylated

pectin. Protopectinase has been regarded as the same enzyme (or enzyme comples) as "pectinase" although the recent works suggest that two enzymes are involved.

Hulme (68) in his review provided the evidence which demonstrates the presence of pectic enzymes in fruits. A high P.G. activity in ripe Bartlett pear has also been reported by McCready and McComb (105). Hulme further reported that the pear pulp at picks made on June 9, June 18 and July 9 shows no protopectinase activity but appreciable P.G. activity. Thereafter unit harvest, no (pectic) enzyme activity is apparent. In fruit ripened off the tree, and in fruit cold stored and then ripened, protopectinase and P.G. activity are clearly present, while in fruit taken directly from store (and not ripened) only slight P.G. activity appears.

Hulme (68) further stated that the failure of attempts to prove the presence of "pectinase" in fruits has been suggested as due to the presence of two inhibitors of this enzyme. It has been suggested that one of these (a thermolabile substance) appears in early August and is responsible for the failure to show P.G. activity from August until harvest. The thermolabile factor appears later and its action could in no way be attributed to the presence of "pectase." The inhibitor is present in the sap of several varieties of pears, and it is not found to have proteolytic character. Although it has been shown that active "pectinase" appears again at a critical period during the normal ripening of pears, inhibitor studies do not make it clear whether this is due to a decrease in the amount of inhibitor present or whether the "pectinase" content is so high at this stage as to be in excess of inhibitor.

It has also been found that inhibition of a given amount of enzyme increases with increasing amount of inhibitor up to a maximum after which no further increase in inhibition is found. This property suggests that "pectinase" may be a mixture of several enzymes.

Pollard and Kieser (122) suggested that "pectase" activity of mature apples varies considerably with variety. It has also been shown that ripe fruits contain, in general, more enzyme than unripe fruits.

Jones and Reid (77) and Demain and Phaff (28) have provided valuable clues to the mode of action of polygalacturonase. Jermyn and Tomkins (148), employing paper chromatographic techniques, have shown that the typical properties of pectin solutions disappear somewhere between an average (polygalacturonic acid) chain length of 32 and 5 galacturonic acid units. They reported that enzymatic degradation of polygalacturonic acid takes place by random scission of the component units in a manner similar to the hydrolysis of cellulose. "Perhaps, following up a suggestion of Date and Hansen (26) and by analogy with other high-molecular metabolic constituents of plants, phosphorylating mechanisms involving nucleotides are concerned."

Nitrogen Compounds (Amino Acids and Protein):

1. Nitrogenous compound found in fruit:

Hulme (68) stated that it has been only in comparatively recent years that serious attempts have been made to link a high nitrogen content of the fruit itself with keeping quality and further to ascertain which component of the nitrogen fraction is responsible for a "nitrogen effect." Various nitrogenous fractions have been found in apples and pears. The total nitrogen content of mature apples and pears is extremely low (less than 80 mg/100 g

fresh weight). In the early, rapid stages of growth, the total nitrogen content may be nearly 300 mg in the pulp and 400 mg in the peel tissue. Of this, protein nitrogen may comprise 30-50 percent in the pulp and 80-90 percent in the more actively metabolizing peel tissue, depending on the stage of development. Hulme and Smith (70) showed that the protein nitrogen per cell varies between 2 and 10 mg x 10-7. They assumed that this protein nitrogen consists of the cytoplasmic lining of the cell. Hulme (68) further suggests that this protein nitrogen represents virtually the total enzyme content of the fruit.

David et al. (27) found some oxidase activity in protein preparations, and suggested that the presence of phosphorus in the ash of preparatiions indicates the presence of nucleoprotein, but in small quantity.

Hulme (61) examined the protein of young Bramley's Seedling apples, and suggested the presence of different amino acids, such as: aspartic and glutamic acids, leucine (and/or isoleucine), serine, glycine, threonine, -alanine, proline, and tyrosline, valine, phenylalanine, arginine, and lysine. McKee and Urbach (106) reported the presence of hydroxproline in the protein of Granny Smith apples. Lysine, phenylalanine, and leucine are the most abundant acids in the "protein" of mature pears.

Hulme (57), using the older, classic methods for the determination of amino and amide nitrogen, found that, in the pulp of apples, more than 50 percent of the nitrogen could be present in a soluble nonprotein form. At certain stages of growth, asparagine accounted for 80 percent, of this alcohol soluble nitrogen. By maturity, asparagine falls down so that at this stage half the soluble nitrogen could be accounted for by amino acids

other than asparagine. Oland (115) considers asparagine to be the main nitrogenous "storage" compound in apple trees as a whole.

The provisional identification of some of the individual amino acids in apples and pears has been first achieved by paper chromatographic methods. Joslyn and Stepka (78) found asparagine, aspartic acid, serine, and y-aminobutyric acid from extracts of Newton Pippin apples; and asparagine, serine, glycine, and a trace of valine from Bartlett pears. Hulme and Arthington (69) showed the presence of y-aminobutyric acid in young fruits of Bramley's Seedling apples. B-Alanine, glycine valine, serine, leucine, tryptophan, gluamic and aspartic acids, phenylalanine, asparagine, and proline were also identified. Piperidine-2-carboxylic acid (pipecolic acid), y-methylproline, homoserine, methylhydroxproline were also identified, but in small quantities. Burroughs (16) working on apples and pears, found that asparagine, aspartic, and glutamic acids are the principal amino acids in these fruits, while moderate amounts of serine, -alanine, y-aminobutyric acid, valine, isoleucine, and methylhydroxyproline are also found. He also noted the traces of peptides in apple juices. According to Elliott (29), an appreciable change in the amino acid pattern during the development of the fruit is in glutamine which is especially conspicuous in very young and in overmature fruits. The absence of glutamine during the intermediate stages of growth may be regarded due to a storage of adenosine triphosphate known to be required for its synthesis in Vitro.

Hulme (68), in his review, reported the presence of relatively large amounts of aspartic acid and asparagine throughout the growth of Williams and Passe-Crassane pears. Smaller amounts of glutamic acid, serine,

threonine, -alanine, valine, and lencine are also present. Lysine is found only at maturity, and phenylalanine is present only in very small quantity. Proline is the most prominant acid at maturity. Burroughs (16), working on Perry pear identified hydroxperidine-2-carboxylic acid. He also isolated 1-aminocyclopropane -1-carboxylic acid.

2. Changes of Nitrogenous Compounds:

A. Changes due to climacteric rise:

There is a special phase in the life history of apples and pears which appears to be associated with the change over from development to senescence. This is the climacteric rise in respiration "the climacteric," a rapid rise in the respiration rate of fruit, which occurs just before visible ripening sets in. During this period the balance between protein and non-protein nitrogen is shifted in favor of protein.

Kidd and West (81) showed that the "climacteric" has a high temperature coefficient and attributed the onset of the climacteric to a change of state in the protoplasm. Hulme (58) found that coinciding with the climacteric rise in respiration in detached apples, there is a rise in the net protein content of the fruit, but there is not change in total nitrogen. Hulme (64) working in different varieties of apples showed that there is always a shift in the equilibrium between non-protein and protein nitrogen in favor of protein over the period of the climacteric. He also showed (66), for fruits picked at any stage of development and stored at 12°C, that fruits which have a climacteric, also exhibit at the same period in their life history a net increase in protein. This also applies to fruits commencing their climacteric on the tree.

B. Changes at different stages of growth:

Archbold (1), Askew (3), Hulme (57), and Robertson and Turner (125) working on several varieties of apples grown in different counties, all found the same pattern of change in the total nitrogen content of the fruits during development on the tree. The concentration of nitrogen is high in the young fruits (a few days after petal fall, as high as 0.35 percent of the fresh weight) falling to as little as 0.02 percent of the fresh weight at maturity. Nevertheless, as long as the fruit increases in weight, the amount of nitrogen per fruit continues to rise. Hulme (125), Robertson and Turner (125) studied the changes in the protein and non-protein nitrogen of the apple during growth. The equilibrium between soluble and protein nitrogen moves rapidly in the direction of soluble nitrogen during the first 60 days from petal fall then move slowly up to 140 days by which time protein nitrogen forms only about 45 percent of the total nitrogen. Thereafter protein nitrogen increases rapidly at the expense of soluble nitrogen until the fruit is harvested.

C. Changes in storage:

Hulme (59) stated that there is no evidence for any significant change in the total nitrogen of apples or pears as for any transfer between pulp and seeds when once they have been detached from the tree. Apart from the changes in the balance between protein and soluble nitrogen which occur over the region of the respiration climacteric, changes in storage are small. It has been found that the pattern of changes in the nitrogen constituents during storage is the same in pear as in apple.

Lipids:

Ulrich (152) has reported several studies that have been made of the lipids from the peel of apples and pears. Oil, wax, ursolic acid and cutin have been extracted from the epidermis of apple peelings. It has been found that the oil fraction of the natural coating increases during storage to a maximum - three to four times the initial value; this increase is less important in gas storage. The oil content also increases with the maturity of the fruit at picking time. The iodine number of the oil increases with the oil concentration. Small increases occur in the wax, ursolic acid and cutin fractions after prolonged storage. The presence of stearic, archidic acids has been found by the method of distillation, with smaller amounts of palmitic and behenic acids and acids of higher molecular weight - tetracosanoic acid for example.

Different Pigments:

A. Pigments other than Flavonoid compounds:

Ulrich (152) says that little systematic work appears to have been carried out on the changes in the carotenoid and chlorophyll in apples and pears. The results of several studies reported have shown the presence of B-carotene, lycopene, tetrahydrolycopene, hydro carbons of the phytofluence series, xanthophylls, cryptoxanthin and chlorophyll in pears and apples. Xanthophylls in fruit differ from the leaf xanthophylls by the fact that they occur mainly as esters. According to Hulme (68) the peel of most deciduous fruits contains many times more of these pigments than does the flesh.

B. Effects of different factors on carotenoid content:

Ulrich (152) reported the effect of light and temperature on the

carotenoids in fruits. A better color in tomato fruits picked at a green stage and ripened at 21°C under light than in those ripened in the dark has been observed. Red light has been found to be particularly active. Better color is obtained for fruits ripened at 16°C than for fruits ripened at 4°C. It has been also found that in green fruits chlorophyll does not disappear and the carotenoids fail to develop in the absence of oxygen unless the fruits are illuminated. In tomato fruits the synthesis of lycopene at 37°C is inhibited without an increase of phytofluene, S-carotene, or neurosporene content. Synthesis of - carotene and B-carotene, is very slightly inhibited at 37°C. At 0°C all synthesis are inhibited. It has also been found that storage temperature has no effect in the changes of color of pear.

C. Evolution of carotenoids during ripening:

According to Ulrich (152) ripening is marked by an increase in the amount of carotenoids in the fruit. The general yellowing of the flesh of apples after removal from the store is due mainly to an increase in the carotene rather than in the carotenol content, and that chlorophyll decreases during ripening. The failure of the skin of pears to yellow during ripening at various temperatures is due to incomplete decomposition of chlorophyll which tends to mark the carotenoids present. Chlorophyll distribution could be hastened by treating the fruit eith ethylene.

He also stated that xanthophylls originate from hydrocarbons by oxidation and that B-methylcrotonaldehyde is the precursor of carotenoids. The different hydrocarbons may arise on out of the other or from a common precursor. Carotenes could arise from colorless polyenes by dehydration,

ring formation, and oxidation. It is also possible that carotenoids might come from ethylene.

Phenolic Constituents:

Hulme (68) stated that phenolic substances are, as the name implies, substances ultimately related to phenol, but the numbers of the group so far identified in the fruits of apple and pear are almost all of a flavonoid nature. Among these components flavones, anthocyanins and catechin derivatives have been recently investigated in fruits. Four leucoanthocyanins are present in various varieties of apples. Chlorogenic acid, and epicatechin also occur in the fruits. Idaein, quercetin, and hyperin have been found as coloring matters in the skin of different varieties of apples. Other compounds such as quercitrin, isoquercitrin, avicularin, rutin, and quercetin 3-xyloside have also been reported.

He also stated that sincemmuch of the astringency, the color of the skin, and the browning of the cut surfaces of apples and pears is due to the presence of phenolic substances, a measure of the total phenolic substances present in the fruit has been a great interest to pomologists and fruit juice cider makers. Because many of these compounds were first recognized in tanning liquors, the name tannin is coined as a generic name. The tannins of pears can be divided into three groups as follows:

1. True tannins - bodies having the properties of tannins and precipitated by gelatin 1 percent aqueous solutions.
2. Nontannin polyphenols - not precipitated by gelatin: compounds such as gallic and ellagic acids.
3. Colored compounds belonging to the class of anthocyanins and flavons.

Vitamin C (Ascorbic Acid):

Hulme (68) reported that fruits are one of the main dietary sources of ascorbic acid; it is not surprising that the ascorbic acid content of apples, and to a less extent pears which have a much lower content of ascorbic acid, has received much attention throughout the world. The ascorbic acid content of apples and pears changes during ripening. This variation may be explained in part by a difference in the maturity of apples and pears of a given variety. The peel from the apple contains three to five times as much ascorbic acid as pulp. Losses of the vitamin during storage at $0^{\circ}C$ and $3^{\circ}C$ are smaller, especially in the peel. A higher content of ascorbic acid in apples exposed to high light intensity has been observed as compared with the inner, shaded fruits on the tree.

He also stated that during the development of apples and pears on the tree and -ascorbic acid content of the whole fruit is highest during the early stages of development when the seeds are still soft and undeveloped, after which it falls rapidly. In general summer apples have a higher concentration of ascorbic acid than fall and winter varieties but they lose the vitamin faster in common storage. The loss of ascorbic acid is somewhat higher in the gas-stored than in the ordinary cold stored fruits, but this may have been a question of temperature difference.

He further stated that low temperature appears to favor synthesis of ascorbic acid, and it is speculated that synthesis may result from the slow liberation of galacturonic acid during the breakdown of pectic substances which occurs in stored fruits. In apples and pears when first placed in storage, ascorbic acid synthesis may be limited by precursors which are subsequently liberated as the tissue softens. The loss of ascorbic acid in

apples and pears stored at higher temperatures might be due, either to the rate of hydrolysis exceeding the rate of synthesis at these temperatures, or to the general slowing down of synthesis which occurs in overripe fruit.

A. Role played by ascorbic acid in the physiology of fruits:

Hulme (68) says that little is known of the role played by vitamins in the physiology of fruits. Vitamin C is found in variable proportions of ascorbic acid (reduced form) and dehydroascorbic acid (oxidized form) in the fruits. The system ascorbic-acid-ascorbic-acid-oxidase-dehydroascorbic acid is probably "geared-in" with the other oxidation-reduction system present in the tissue such as the cytochromes and cytochrome oxidase and the polyphenols and polyphenoloxidase. There is, however, little positive evidance one way or another that ascorbic acid acts as a respiratory catalyst. Ascorbic acid may also be concerned in oxidations and reductions involving glutathione. Polyphenolase activity closely parallels Vitamin C oxidation capacity except in ripe pears.

Auxins:

The hormones controlling growth have become known generally as the auxins i.e., growth regulators which induce cell enlargement at low concentrations or the substances which affect extension of the cell wall, accompanied by water uptake in the cell. Modern techniques in plant hormone analysis, in particular the use of paper chromatography for the purification of extracts, followed by bioassay, have revealed a number of as yet chemically unidentified auxins in various plant species. The fruit auxins are mainly localized in the seeds.

Nitsch (114) has made an important report on the presence and role of

auxins in the growth fruit. Among the various workers Luckwill (96) and Wright (161) have reported the role of auxins in relation to fruit growth and fruit drop. Ulrich stated that very little work has been done on the role of auxins in postharvest fruits. However, the end of fruit growth and maturity are marked by a very low auxin content. During storage the auxin content remains very low and inhibitors are present.

CONCLUSION

In spite of the fact that several papers have been published since 1950, it is not yet possible to give a comprehensive view of the life of postharvest fruit. Ulrich (152) says: "The end of the life of a fruit is generally a slow agony, but it is often preceded by a period of great activity although growth is over. Ripening is characterized by intense oxidations, in which mitochondria play an important role; by the appearance or increase of certain constituents such as pigments, volatiles, ethylene, sucrose, soluble pectins; and finally by the decrease of other constituents such as organic acids, chlorophyll and auxins."

Though the efforts have been made to find some links between the different physiological reactions - such as between ethylene evolution and respiration, pectin production and respiration, between the different acids or carotenes, between acidity or ascorbic acid and pectin; yet new experiments are necessary.

The effects on the storage life of the apple and pear on the nutritional factors, climate and soil, and of a number of other prestorage conditions have been determined but a more extensive study of the effects of these variables

and of their interrelationships is very much required.

LITERATURE CITED

1. Archbold, H. K. 1932.
Chemical studies in the physiology of apples. XII. Ripening process in the apple and the relation of time of gathering to the chemical changes in cold storage. Ann. Botany (London) 46:407.

2. Ash, A. S. F., and Reynolds, T. M. 1955.
Water soluble constituents of fruits. III. An examination of the sugars and polyols of apricots, peaches, pears and apples by paper chromatography. Australian J. Chem. 8:276.

3. Askew, H. O. 1935.
Changes in the chemical composition of developing apples. J. Pomol. Hort. Sci. 13:232.

4. Axelord, B., and Seegmiller, C. G. 1954.
Metabolism of apples. Conversion of glucose to sucrose in apple tissues. J. Agr. Food Chem. 2:1329.

5. Baker, C. E. and Maxie, E. C. 1952.
An apparent relation of a physiological spot on Red Rome apples in storage by activated charcoal and shredded oil paper. Proc. Am. Soc. Hort. Sci. 59:312-314.

6. Bartholomew, E. T., and Sinclair, W. B. 1951.
The lemon fruit. University of California Press, Berkley and Los Angeles.

7. Biale, J. B. 1950.
Postharvest physiology and biochemistry of fruits. Ann. Rev. Plant Physiol. 1:183-206.

8. Biale, J. B., Young, R. E., and Olmstead, A. J. 1954.
Fruit respiration and ethylene production. Plant Physiol. 29:168-174.

9. Biale, J. B., Young, R. E., Popper, C. S., and Appleman, W. E. 1957.
Metabolic processes in cytoplasmic particles of the avocado fruits. I. Preparative procedure, co-factors requirements and oxidative phorphorylation. Physiol. Plant 10:48-63.

10. Blake, C. E., and Shirley, R. L. 1953.
Oxidation potentials, buffering, ash, and total solids of peaches. Bot. Gaz. 115:180-185.

11. Bonner, J. 1950.
Plant Biochemistry. Academic Press New York.

12. Brooks, C. 1935.
Some botanical aspects of perishable food products. Sci. Monthly 40:122-137.

13. Brooks, C., and Cooley, J. S. 1917.
Effect of temperature, aeration, and humidity on Jonathan-spot and scald of apples in storage. J. Agr. Research 11: 287-313.

14. Brooks, C., Cooley, J. S., and Fisher, D. F. 1919.
Nature and control of apple scald. J. Agr. Research 18: 211-240.

15. Brooks, C., Cooley, J. S., and Fisher, D. F. 1923.
Oiled wrappers, oils and waxes in the control of apple scald. J. Agr. Research 25:513-536.

16. Burrough, L. F. 1957.
1-amino-cyclopropane 1-carboxylic acid, a new amino acid in Perry pears and cider apples. Nature 179:360.

17. Caldwell, J. 1956.
Studies in respiration of apple at various pressure of oxygen. J. Exptl. Botany 7:326-334.

18. Chanda, S. K., Hirst, E. L., and Perceval, E. G. V. 1951.
The constitution of pear cell-wall xylan. J. Chem. Soc. p. 1240.

19. Cheng-Tan Chow, and Biale, J. B. 1953.
Metabolic processes in cytoplasmic particles of the avocado fruit. II. Participation of cytochrome C in the electron transport chain. Physiol. Plant. 10:54-75.

20. Childers, N. F. 1954.
Modern fruit science. Horticultural Publications. Rutgers, the State University, New Jersey.

21. Claypool, L. L., and Allen, F. W. 1951.
The influence of temperature and oxygen level on the respiration and ripening of Wickson plums. Hilgardia 21:129-160.

22. Claypool, L. L., Maxie, E. C., and Esau, K. 1955.
Effect of aeration rate on the respiratory activity of some deciduous fruits. Proc. Am. Soc. Hort. Sci. 66:125-134.

23. Christensen, B. E., Hansen, E., and Cheldelin, V. H. 1939.
Determination of ethylene contained in the internal atmosphere of the plant tissues. Ind. and Eng. Chem. Anal. Ed. II:114-116.

24. Cummings, M. B., and Lombard, P. M. 1915.
Farm apple storage. Vt. Agr. Expt. Sta. Bull. 186:99-136.

25. Curtis, O. F. 1937.
Vapor pressure gradients, water distribution in fruits and so-called infra-red injury. Am. Jour. Botany 24: 705-710.

26. Date, W. B., and Hansen, E. 1954.
Pectin changes in pears during storage and ripening. Proc. Indian Acad. Sci. (B) 39:17.

27. Davis, S. G., Fillers, C. R., and Esselen, W. B. 1949.
Composition and nature of apple protein. Food Research 14:417.

28. Demain, A. L., and Phaff, H. J. 1954.
Hydrolysis of the oligo-galacturonides and pectin acid by yeast polygalacturonase. J. Biol. Chem. 210:381.

29. Elliott, W. H. 1948.
Adenosinetriphosphate in glutamine synthesis. Nature 161:128.

30. Filder, J. C. 1950.
Studies of the physiologically active volatile organic compounds produced by fruits. III. The rate of production of carbon dioxide and of volatile organic compounds by King-Edward VII apples in gas storage, and the effect of removal of volatiles from the atmosphere of the store on the incidence of superficial scald. J. Hort. Sci. 25:81-110.

31. Filder, J. C. 1950.
A comparison of the aerobic and anaerobic respiration of apples. J. Exptl. Botany 2:41-64.

32. Filder, J. C. 1955.
Volatile organic products of metabolism of fruits. J. Sci. Food Agr. 6:293-295.

33. Fisher, D. V., and Porritt, S. W. 1955.
Some recent studies in late harvesting and delayed cold storage of Bartlett pears. Proc. Am. Soc. Hort. Sci. 65: 223-230.

34. Gane, R. 1935.
Formation of ethylene by plant tissues and its significance in the ripening of fruits. J. Pomol. Hort. Sci. 13:351.

35. Gerhard, F. G. 1954.
Rates of emanation of volatiles from pears and apples. Pro. Am. Soc. Hort. Sci. 64:248-254.

36. Gerhard, F. G., and Allmendiger, D. F. 1945.
Influence of a-naphthaleneacetic acid spray on maturity and storage physiology of apples, pears and sweet cherries. Jour. Agr. Research 73:189-206.

37. Graubard, M., and Nelson, J. M. 1936.
On the quantity measurement of the enzyme Tyrosinase. J. Biol. Chem. 112:135-141.

38. Grierson-Jackson, W. R. F. 1952.
Cooling methods affect volatile content in storage atmosphere. Refrig. Eng. 60(2):148-152.

39. Griffiths, D. G., and Potter, N. A. 1945.
The determination of starch in apple tissue. Biochem. J. 39:423.

40. Griffiths, D. G., and Potter, N. A. 1949.
Effect of the accumulation of the volatile substances produced by apples in gas storage. J. Hort. Sci. 25: 10-18.

41. Griffiths, D. G., and Potter, N. A. 1950.
Effects of ethylene on respiratory activity of apples in gas storage, with special reference to stage of maturity. J. Hort. Sci. 26:1-7.

42. Griffiths, D. G., Potter, N. A., and Kulme, A. C. 1950.
Data for the study of the metabolism of apples during growth and storage. J. Hort. Sci. 25:266.

43. Hackney, F. M. V. 1948.
Studies in the metabolism of apple. VII. Study of the Polyphenolase System in apples. Proc. Linn. Soc. N.S.W. 73:439.

44. Hall, W. C. 1951.
Studies on the origin of ethylene from plant tissues. Bot. Gaz. 113:55-65.

45. Haller, M. H. 1929.
Pectic constituents in ripe and unripe fruits. J. Agr. Research 39:739.

46. Haller, M. H., and Lutz, J. M. 1941.
Apple storage losses at 36°F. U. S. Dept. Agr. Tech. Bull. 776.

47. Hansen, E. 1942.
Quantitative study of ethylene production in relation to respiration of pears. Bot. Gaz. 103:543-558.

48. _____. 1943.
Relation of ethylene production to respiration and ripening of premature pears. Proc. Am. Soc. Hort. Sci. 43:69-72.

49. _____. 1945.
Quantitative study of ethylene production in apple varieties. Plant. Physiol. 20:631-635.

50. _____. 1946.
Effect of 2,4-Dichlorophenoxyacetic acid on the ripening of Bartlett pears. Plant. Physiol. 21:588-592.

51. Hansen, E., and Christensen, B. E. 1940.
Chemical determination of ethylene in the emanation of apples and pears. Bot. Gaz. 101:403-409.

52. Hartman, H. 1931.
A preliminary report on Anjou scald and its control. Org. Agr. Expt. Sta. Bull. 280.

53. Hawkins, L. A. 1922.
The effect of low-temperature storage and freezing on fruits and vegetables. Am. Jour. Botan. 9:551-556.

54. Haynes, D. 1925.
Chemical studies in the physiology of apples. I. The change in the acid content of stored apples and its physiological significance. Ann. Botany (London) 39:77.

55. Heinze, R. E., Baker, C. E., and Quackenbush, F. W. 1953.
The chemical composition of apple storage volatiles. I. Acids, alcohols, and esters. Proc. Am. Soc. Hort. Sci. 61:237-745.

56. Huber, W., Brasch, A., and Waly, A. 1952.
Effect of processing conditions on organoleptic changes in foodstuffs sterilized with high intensity electrons. Food Tech. 7:109-115.

57. Hulme, A. C. 1936.
Biochemical studies in the nitrogen metabolism of the apple fruit. II. The course followed by certain nitrogen fractions during development of the fruit on the tree. Biochem. J. 30:258.

58. _____. 1937.
Changes in the nitrogenous compounds of the apple in the period of the climacteric rise in respiratory activity. Rept. Food Invest. Board, London, 1936. p. 128.

59. _____. 1948.
Studies in the nitrogen metabolism of apple fruit. Changes in the nitrogen metabolism of the apple during the normal ethylene induced climacteric rise in the rate of respiration. Biochem. J. 43:343.

60. _____. 1950.
Data for the study of the metabolism of apples during growth and storage. Section II. J. Hort. Sci. 25:277.

61. _____. 1951.
The amino acids present in the protein of apple fruit. J. Sci. Food Agr. 2:161.

62. _____. 1951.
The isolation of l-quinic acid from the apple fruit. J. Exptl. Botany 2:298.

63. _____. 1953.
The isolation of chlorogenic acid from the apple fruit. Biochem. J. 53:337-340.

64. _____. 1954.
The relation between the rate of respiration of an apple fruit and its content of protein. II. The value of the relation immediately after picking and at the respiration-climacteric for several varieties of apples. J. Hort. Sci. 29:98-103.

65. _____. 1954.
The oxidation of citramalic acid and B-hydroxyglutaric acid to acetoacetic acid and the possible significance of the citramalic acid in plant metabolism. Biochem. et. Biophys. Acta. 14:44.

66. _____. 1954.
Studies in the nitrogen metabolism of apple fruits. The climacteric rise in respiration in relation to the equilibrium between protein synthesis and breakdown. J. Expt. Botany 5:159.

67. _____. 1956.
Carbon dioxide injury and the presence of succinic acid in apple. Nature 178:218-219.

68. _____. 1958.
Biochemistry of apple and pear fruits. Advances in Food Research. 8:297-413.

69. Hulme, A. C., and Arthington, W. 1950.
β-amino butyric acid and Y-alanine in plant tissue. Amino acids of the apple fruit. Nature 165:716.

70. Hulme, A. C., and Smith, W. H. 1939.
A relation between protein content and respiration in the cell of the apple. Rept. Food Invest. Board, London, p. 127.

71. Ingraham, L. L. 1956.
Effect of ascorbic acid on polyphenol oxidase. J. Am. Chem. Soc. 78:5095-5097.

72. James, W. O. 1953.
The terminal oxidases of plant respiration. Biol. Rev. 28: 245-260.

73. James, W. O., and Beevers, H. 1950.
The respiration of Arum spadix. A rapid respiration resistant to cyanide. New Phytol. 49:353.

74. Jansen, E. F., and MacDonell, L. R. 1945.
Influence of the methoxyl content of pectic substances on the action of polygalacturonase. Arch. Biochem. 8:97.

75. Jermy, M. A., and Isherwood, F. A. 1956.
Changes in the cell wall of the pear during ripening. Biochem. J. 64:123.

76. Jonathan, W. White, Jr. 1949.
Composition of a volatile fraction of apples. Food Research 15:68-78.

77. Jones, J. K. N., and Reid, W. W. 1954.
The structure of the oligosaccharides produced by the enzymic breakdown of pectic acid. Part I. J. Chem. Soc. p. 1361.

78. Joslyn, M. A., and Stepka, W. 1949.
The free amino acids of fruits. Food Research 14:459.

79. Joslyn, M. A., and Ponting, J. D. 1951.
Enzyme-catalyzed oxidative browning of fruit products. Advances in Food Research 3:1.

80. Kertesz, Z. I. 1951.
The pectic substances. Interscience Publishers, Inc. New York, 1951.

81. Kidd, F., and West, C. 1930.
Physiology of fruit. Part I. Changes in the respiratory activity of apples during their senescence at different temperatures. Proc. Roy. Soc. B, 106:93.

82. Kidd, F., and Hansen, C. F. 1937.
Hydrogen ion concentration in apples. Rept. Food Invest. Board, London, 1936, p. 133.

83. Kidd, F., and West, C. 1938.
The effect of ethylene on the respiration activity and the climacteric of apples. Rept. Food Invest. Board, London, 1937, pp. 108-114.

84. _____. 1938.
Spotting and other effects on apples in storage due to volatile products from ripe apples of other varieties stored with them. J. Hort. Sci. 16:274-279.

85. Kidd, F., West, C., Griffiths, D. G., and Potter, N. A. 1940.
An investigation of the chemical changes of Conference pears. Ann. Botany (London) 4:1.

86. _____. 1950.
The degradation of starch in apples removed from the tree at different stages of development. J. Hort. Sci. 25:289-296.

87. _____. 1951.
Metabolism of malic acid in apples. J. Hort. Sci. 26:169.

88. Kidd, F., and West, C. 1952.
The storage qualities of late dessert varieties of apples. Food Invest. Board, London, Tech. Paper No. 2.

89. Kidd, F., West, C., Griffiths, D. G., and Potter, N. A. 1952.
Metabolism of sucrose in apples. I. J. Hort. Sci. 27:179.

90. Kieser, M. E., and Pollard, A. 1951.
The effect of fruit storage on apple juice processing. II. Ann. Rept. Agr. Hort. Res. Sta. Long Ashton, Bristol, 1951, p. 188.

91. Krotkov, G., and Nelson, V. 1946. Carbohydrate metabolism of McIntosh apples during their development on tree and in cold storage. Can. J. Research (C) 24:126.

92. Krotkov, G., Wilson, D. G., and Street, R. W. 1951.
Acid metabolism of the McIntosh apples during their development on tree and in cold storage. Can. J. Botany 29:79.

93. Kuc, J., Heinze, R. E., and Quackenbush, F. W. 1953.
Apple scald. Production and control in the laboratory. J. Agr. Food Chem. 1:1104-1107.

94. _____. 1953.
Apple scald. Use of alkaline permangnet for control in refrigerated storage. J. Sgr. Food Chem. 1:1107-1109.

95. Leonard, S., Luh, B. S., Hinreiner, E., and Simone, M. 1954.
Maturity of Bartlett pears for canning. Food Tech. 8:478-482.

96. Luckwill, L. C. 1953.
Studies on fruit development in relation to plant hormones. I. Hormone production by the developing apple seed in relation to fruit drop. J. Hort. Sci. 28:14.

97. Magness, J. R. 1930.
Composition of gases in intercellular spaces of apples and potatoes. Bot. Gaz. 70:308.

98. Magness, J. R. et al. 1926.
The ripening, storage, and handling of apples. U. S. Dept. Agr. Bull. 1406.

99. Marshall, R. E. 1954. Cherries and cherry products. Interscience Publishers Inc., New York.

100. Marshall, R. E. et al. 1936.
The relation of washing treatment to subsequent losses in moisture from apples. Wash. Agr. Expt. Sta. Bull. 330:1-28.

101. Martin, W. E. 1937.
Chemical study of ripening process of Bose Pears. Bot. Gaz. 99:42.

102. Martin, C. M., and Reuter, F. H. 1949.
Nature 164:373.

103. Mattus, G. E. 1950.
Bartlett pear respiration and volatile production after storage in air vs. in controller atmosphere. Proc. Am. Soc. Hort. Sci. 55:199.

104. Maxie, E. C., and Baker C. E. 1955.
Air filtration studies in a commercial type apple storage. Proc. Am. Soc. Hort. Sci. 64:235-247.

105. McCready, R. M., and McComb, E. A. 1954.
Pectic substances in ripe and unripe fruits. Food Research 19:530-535.

106. McKee, H. S., and Urbach, G. E. 1953
The physiology of growth in apples fruits. V. Soluble nitrogen constituents. Australian J. Biol. Sci. 6:369.

107. Meigh, D. F. 1956.
Volatile compounds produced by apples. I. Aldehyde and Ketones. J. Sci. Food Sgr. 6:396.

108. Meigh, D. F. 1957.
Volatile compounds produced by apples. II. Alcohols and Esters. J. Sci. Food Agr. 8:313-325.

109. Miller, E. V. 1946.
Physiology of citrus fruits in storage. Bot. Rev. 12:393-423.

110. Mitchell, J. W., and Marth, P. C. 1944.
Effects of 2,4-Dichlorophenozyacetic acid on the ripening of detached fruits. Bot. Gaz. 106:199-207.

111. Morgan, B. H. 1955.
Food radiation roundup--how different foods stand up. Food Eng. 27:44.

112. Nelson, R. C. 1937.
The quantity of ethylene present in apples. Plant Physiol. 12:1004-1005.

113. _____. 1939.
Studies on the production of ethylene in the ripening process of apple and banana. Food Research 4:173-190.

114. Nitsch, J. P. 1953.
The physiology of fruit growth. Ann. Rev. Plant Physiol. 4:199-236.

115. Oland, K. 1955.
Two field trials with different nitrogen fertilization of the apple variety Gravenstein. Ullenswarg Research Sta. Loftus, Norway, Rept. No. 7.

116. Onslow, M. W. 1920.
Oxidizing enzymes. III. Oxidizing enzymes of some common fruits. Biochem. J. 14:541-547.

117. Onslow, M. W., Kidd, F., and West, C. 1932.
Biochemical study of senescence in apples. I. Comparison of chemical change in apples from different localities. Rept. Food Invest. Board, London, 1931, p. 52.

118. _____. 1933.
Biochemical study of senescence in apples. Rept. Food Invest. Board, London, 1932, p. 70.

119. Pentzer, W. T., and Heinze, P. H. 1954.
Postharvest physiology of fruits and vegetables. Ann. Rev. Plant Physiol. 5:205-224.

120. Pieniazek, S. 1943.
Maturity of apple fruits in relation to the rate of transpiration. Proc. Am. Soc. Hort. Sci. 42:231-237.

121. Poast, P. A., and Phillips, W. R. 1952.
Air purification and soluble pectin accumulation in Lawfan apple. Sci. Agr. 32:109.

122. Pollard, A., and Kieser, M. E. 1951.
The pectase activity of apples. J. Sci. Food Agr. 2:30.

123. Powell, G. H., and Fulton, S. H. 1903.
The apple in cold storage. U. S. Dept. Agr. Bur. Plant Ind. Bull. 48.

124. Reeve, R. M. 1953.
Histological investigation of texture in apples. II. Structure and intercellular spaces. Food Research 18: 604-617.

125. Robersons, R. N., and Turner, J. F. 1951.
The physiology of growth of apple fruits. II. Respiration and other metabolic activities as function of cell number and cell size in fruit development. Australian J. Sci. Research (B) 4:92.

126. Siegelman, H. W. 1954.
The sugars of Grimes Golden apple and Bartlett pear skin. Proc. Am. Soc. Hort. Sci. 64:232.

127. _____. 1955.
Polyphenol oxidase substrates of apple and Bartlett pear skin. Arch. Biochem. Biophys. 65:97-102.

128. Shutak, V., and Christopher, E. P. 1953.
Effect of mineral oil storage scald apples. Proc. Am. Soc. Hort. Sci. 63:233-236.

129. Smith, W. H. 1931.
Loss of water from fruit. (Gr. Br.) Rept. Sci. & Ind. Res., Food Invest. Board, Rep. 1930:55-56.

130. _____. 1932.
Evaporation from apples. (Gr. Br.) Rept. Sci. & Ind. Res., Food Invest. Board, Rep. 131:152-153.

131. Smock, R. M. 1943.
The influence of stored apples on the ripening of other apples stored with them. Cornell Univ. Agr. Expt. Sta. Bull. 799.

132. _____. 1944.
The physiology of deciduous fruits in storage. Botan. Rev. 10:560-598.

133. _____. 1955.
Apple volatiles and their significance at storage temperatures. Proc. Am. Soc. Hort. Sci. 66:111-117.

134. _____. 1957.
A comparison of treatments for control of the apple scald disease. Proc. Am. Soc. Hort. Sci. 69:91-100.

135. Smock, R. M., and Southwick, F. W. 1945.
Studies on storage scald of apples. New York (Cornell) Agr. Expt. Sta. Bull. 813.

136. Smock, R. M., and Gross, C. R. 1947.
The effect of some hormone materials on the respiration and softening rates of apples. Proc. Am. Soc. Hort. Sci. 49:67-80.

137. Smock, R. M., and Southwick, F. W. 1948.
Studies on storage scald of apples. New York (Cornell) Agr. Expt. Sta. Bull. 843.

138. Smock, R. M., and Nuebert, A. N. 1950.
Apple and apple products. Interscience Publishers Inc. New York.

139. Smock, R. M., and Gross, C. R. 1952.
The effects of the vapors of different quantities on the ripening rate of apples. Proc. Am. Soc. Hort. Sci. 59: 307-311.

140. Southwick, F. W. 1945.
Removal of organic emanations from apple store rooms. Jour. Agr. Research 71:297.

141. _____. 1946.
Effect of some growth regulating substances on the rate of softening, respiration, and soluble solids content of peaches and apples. Proc. Am. Soc. Hort. Sci. 47:84-90.

142. Southwick, F. W., and Smock, R. M. 1943.
Lengthening the storage life of apples by removal of volatile materials from the storage atmosphere. Plant Physiol. 18:716-717.

143. Speiser, R. 1947.
Advances in pectin chemistry. Part I.

144. Stitt, F., and Tomimastu, Y. 1951.
Sensitizer paper for estimation of mercury vapor. Anal. Chem. 23:1098-1101.

145. Strain, H. 1937.
Sources of d-sorbitol. J. Am. Chem. Soc. 59:2267.

146. Thompson, A. R. 1951.
Volatile products of apples. I. Identification of acids and alcohols. Australian J. Sci. Research (B) 4:283-290.

147. Thompson, A. R., and Huelin, F. E. 1951.
Volatile products of apples. II. Production of volatile esters by Granny Smith apples. Australian J. Sci. Research (B) 4:544-553.

148. Tompkins, R. G. 1954.
Unsolved problems in the preservation of food. The influence of cultural conditions on the quality and preservation of fruits and vegetables. J. Sci. Food Agr. 5:161.

149. Trout, S. A., Hall, E. G., and Sykes, S. M. 1953.
Effects of skin coatings on the behavior of apples in storage. Australian J. Agr. Res. 4:57-81.

150. Turner, J. F. 1949.
The metabolism of apples during storage. Australian J. Sci. Research (B) 2:138.

151. Tutin, F. 1925.
Chemical investigations of fruits and their products. I. Apple juice as a source of sorbitol. Biochem. J. 19:416.

152. Ulrich, R. 1958.
Postharvest physiology of fruits. Ann. Rev. Plant Physiol. 9:385-416.

153. Uota, M. 1955.
Effect of temperature and ethylene on evolution of carbon dioxide, ethylene, and other oxidizable volatiles from three varieties of plums. Proc. Am. Soc. Hort. Sci. 65: 231-243.

154. Uota, M., and Dewy, D. H. 1953.
The respiration and volatile emanation of Bartlett pears as influenced by postharvest treatment with ethylene and 2,4,5-T. Proc. Am. Soc. Hort. Sci. 61:253-264.

155. Watts, J. H., and Griswold, R. M. 1953.
Enzyme and ascorbic acid content of fresh and frozen pine apple. Food Research 18:162-168.

156. Webster, G. C. 1954.
The effect of carbon monoxide on respiration in higher plants. Plant Physiol. 29:399-400.

157. Weurman, C., and Swain, T. 1955.
Changes in the enzymic browning of Bramley's seedling apples during their development. J. Sci. Food. Agr. 6: 180-192.

158. White, J. W. 1950.
Composition of a volatile fraction of apples. Food Research 15:68-78.

159. Widdowson, E. M. 1932.
Chemical studies in the physiology of apples. XIII. The starch and hemicellulose content of developing apples. Ann. Botany (London) 36:597.

160. Wright, R. C. 1939.
Low-temperature effects on the physiology of plant organs in relation to commercial storage. Ice and Refrig. 97: 261-264.

161. Wright, S. T. C. 1956.
Studies of fruit development in relation to plant hormones. III. Auxins in relation to fruit morphogenesis and fruit drop in the black currant (Ribes nigrum). J. Hort. Sci. 31:196-211.

162. Young, R. E., Pratt, H. K., and Biale, J. B. 1952.
Manometric determination of low content of ethylene. Anal. Chem. 24:551-555.

POSTHARVEST PHYSIOLOGY OF POMACEOUS FRUITS

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AN ABSTRACT OF

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ABSTRACT

Several works have been reported on the postharvest physiology of fruits. The purpose of this report is to review the works done on postharvest physiology of pomaceous fruits exclusively. The main new facts and opinions concerning the conditions of ripening, the gas exchange, and the metabolism of postharvest physiology of pomaceous fruits have been reviewed here.

There are various factors which affect the conditions of ripening. These are temperature, humidity, ethylene, volatiles, and growth substances.

Harvested fruits receive oxygen and sometimes water vapor or CO_2 from the environment and give out CO_2, ethylene, volatiles, and water vapor. The marked rise in oxygen uptake and CO_2 output known as "climacteric rise" is a characteristic phenomenon of the ripening process. Respiratory activity is affected by temperature, oxygen concentration, ethylene, and growth substances.

The loss of carbon in the form of volatile organic substances never exceeds 1 percent of that lost as CO_2. In term of carbon, ethylene forms 70-80 percent of the total. The remainder which includes alcohol, esters, and aldehydes and ketones, is referred to as "odorous fraction."

The main substrates in respiration are sugars and organic acids. During the ripening period the oxidation-reduction potential is found to be on the reduction side, while during over-ripeness it is more on the oxidation side. The respiratory activity catalyzed by metal enzymes increases at the same time. The respiratory rate is governed by the amount of ADP (Adenosine diphosphate) which is available to accept phosphate.

Degradation of starch proceeds at the rate proportional to the surface area of the starch grain. The presence of soluble sugars in fruits has been determined. Immediately after picking, sucrose increases only a little, after that it decreases to a low level. Soluble pectin increases during ripening at the expense of protopectin, but the qualitative changes of pectin are not well known.

The total nitrogen content of mature apples and pears is extremely low. Several amino acids have been found in the extract of apple and pear juice and their content varies during the preharvest and postharvest period. Several organic acids have been detected in fruits by new chromatographic methods. Organic acids do not contribute to the increase in sugars during the climacteric and are not formed during the decrease of sugars in the post-climacteric period. Organic acids are formed by the process of carboxylation.

It has been found that the epidermis of apple peelings contain oil, wax, ursolic acid and "cutin." The oil fraction increases to the maximum during storage, with a small increase in wax, ursolic acid and "cutin." The content of oil also increases with the maturity of fruit.

B-carotene is always present in fruits. In the presence of lycopene B-carotene is a minor constituent. Xanthophylls in fruits differ from leaf xanthophylls by the fact that they occur mainly as esters. Ripening is marked by an increase in the amount of carotenoids in fruits. Light, temperature and oxygen affect the amount of carotenoids in fruits.

Among the phenolic constituents flavones, anthocyanins and catechin derivatives have been investigated. The fruit auxins are mainly localized

in the seeds. The end of fruit growth and maturity are marked by a very low auxin content. During storage the auxin content remains very low and inhibitors for auxin are present.